Amino Acid and nucleotide sequence of the murine OKT3 heavy chain variable region (Accession #A222621)

```
MERHWIFLLLLSVTAGVHSQVQLQQSGAELARPGASVKMSCKASYTFTRYTMHWVKQRPGQGLEWIGYINPSRGYTN
YNQKFKDKATLTTDKSSSTAYMQLSSLTSEDSAVYYCARYYDDHYCLDYWGQGTTLTVSSAKTTAPSVYPLAPVCGD
TTGSSVTLGCLVKGYFPEPVTLTWNSGSLSSGVHTFPAVLQSDLYTLSSSVTVTSSTWPSQSITCNVAHPASSTKVD
KKIEPRGPTIKPCPPCKCPAPNLLGGPSVFIFPPKIKDVLMISLSPIVTCVVVDVSEDDPDVQISWFVNNVEVHTAQ
TQTHREDYNSTLRVVSALPIQHQDWMSGKEFKCKVNNKDLPAPIERTISKPKGSVRAPQVYVLPPPEEEMTKKQVTL
TCMVTDFMPEDIYVEWTNNGKTELNYKNTEPVLDSDGSYFMYSKLRVEKKNWVERNSYSCSVVHEGLHNHHTTKSFS
RTPGK

ORIGIN
      1 gaattcccct ctccacagac actgaaaact ctgactcaac atggaaaggc ctggatctt
     61 tctactcctg ttgtcagtaa ctgcaggtgt ccactcccag gtccagctgc agcagtctgg
    121 ggctgaactg gcaagacctg gggcctcagt gaagatgtcc tgcaaggctt ctggctacac
    181 ctttactagg tacacgatgc actgggtaaa acagaggcct ggacagggtc tggaatggat
    241 tggatacatt aatcctagcc gtggttatac taattacaat cagaagttca aggacaaggc
    301 cacattgact acagacaaat cctccagcac agcctacatg caactgagca gcctgacatc
    361 tgaggactct gcagtctatt actgtgcaag atattatgat gatcattact gccttgacta
    421 ctggggccaa ggcaccactc tcacagtctc ctcagccaaa acaacagccc catcggtcta
    481 tccactggcc cctgtgtgtg gagatacaac tggctcctcg gtgactctag gatgcctggt
```

FIG. 1A

```
541 caagggttat ttccctgagc cagtgacctt gacctggaac tctggatccc tgtccagtgg
601 tgtgcacacc ttcccagctg tcctgcagtc tgacctctac accctcagca gctcagtgac
661 tgtaacctcg agcacctggc ccagccagtc catcacctgc aatgtggccc acccggcaag
721 cagcaccaag gtggacaaga aaattgagcc cagagggccc acaatcaagc cctgtcctcc
781 atgcaaatgc ccagcaccta acctcttggg tggaccatcc gtcttcatct tccctccaaa
841 gatcaaggat gtactcatga tctccctgag ccccatagtc acatgtgtgg tggtggatgt
901 gagcgaggat gacccagatg tccagatcag ctggtttgtg aacaacgtgg aagtacacac
961 agctcagaca caaacccata gagaggatta caacagtact ctccgggtgg tcagtgccct
1021 ccccatccag caccaggact ggatgagtgg caaggagttc aaatgcaagg tcaacaacaa
1081 agacctccca gcgcccatcg agagaaccat ctcaaaaccc aaagggtcag taagagctcc
1141 acaggtatat gtcttgcctc caccagaaga agagatgact aagaaacagg tcactctgac
1201 ctgcatggtc acagacttca tgcctgaaga catttacgtg gagtggacca acaacgggaa
1261 aacagagcta aactacaaga acactgaacc agtcctggac tctgatggtt cttacttcat
1321 gtacagcaag ctgagagtgg aaaagaagaa ctgggtggaa agaaatagct actcctgttc
1381 agtggtccac gagggtctgc acaatcacca cacgactaag agcttctccc ggactccggg
1441 taaatgagct cagcacccac aaaactctca ggtccaaaga gacacccaca ctcatctcca
1501 tgcttccctt gtataaataa agcacccagc aatgcctggg accatgtaaa aaaaaaaaaa
1561 aaaggaattc
```

FIG. 1A (Cont.)

Amino Acid and nucleotide sequence of the murine OKT3 light chain variable region (Accession #A22259)

```
MDFQVQIFSFLLISASVIISRGQIVLTQSPAIMSASPGEKVTMTCSASSSVSYMNWYQQKSGTSPKRWIYD
TSKLASGVPAHFRGSGSGTSYSLTISGMEAEDAATYYCQQWSSNPFTFGSGTKLEINRADTAPTVSIFPPS
SEQLTSGGASVVCFLNNFYPKDINVKWKIDGSERQNGVLNSWTDQDSKDSTYSMSSTLTLTKDEYERHNSY
TCEATHKTSTSPIVKSFNRNEC

ORIGIN
      1 gaattcccaa agacaaaatg gattttcaag tgcagatttt cagcttcctg
         ctaatcagtg
     61 cctcagtcat aatatccaga ggacaaattg ttctcaccca gtctccagca
         atcatgtctg
    121 catctccagg ggagaaggtc accatgacct gcagtgccag ctcaagtgta
         agttacatga
    181 actggtacca gcagaagtca ggcacctccc ccaaaagatg gatttatgac
         acatccaaac
    241 tggcttctgg agtccctgct cacttcaggg gcagtgggtc tgggacctct
         tactctctca
    301 caatcagcgg catggaggct gaagatgctg ccacttatta ctgccagcag
         tggagtagta
    361 acccattcac gttcggctcg gggacaaagt tggaaataaa ccgggctgat
         actgcaccaa
    421 ctgtatccat cttcccacca tccagtgagc agttaacatc tggaggtgcc
         tcagtcgtgt
```

FIG. 1B

```
481 gcttcttgaa caacttctac cccaaagaca tcaatgtcaa gtggaagatt
     gatggcagtg
541 aacgacaaaa tggcgtcctg aacagttgga ctgatcagga cagcaaagac
     agcacctaca
601 gcatgagcag caccctcacg ttgaccaagg acgagtatga acgacataac
     agctatacct
661 gtgaggccac tcacaagaca tcaacttcac ccattgtcaa gagcttcaac
     aggaatgagt
721 gttagagaca aaggtcctga gacgccacca ccagctccca gctccatcct
     atcttccctt
781 ctaaggtctt ggaggcttcc ccacaagcgc ttaccactgt tgcggtgctc
     taaacctcct
841 cccacctcct tctcctcctc ctccctttcc ttggctttta tcatgctaat
     atttgcagaa
901 aatattcaat aaagtgagtc tttgccttga aaaaaaaaaa aaa
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FIG. 1B (Cont.)

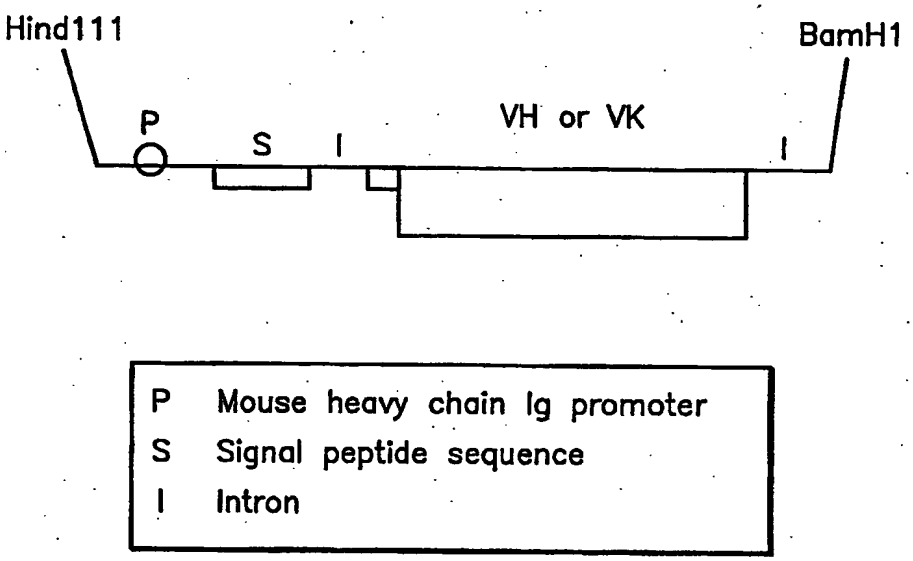

Hind111
BamH1
P
S
I
VH or VK
I
P Mouse heavy chain Ig promoter
S Signal peptide sequence
I Intron

FIG. 2

OKT3 VH gene construct. Nucleic Acid and amino acid sequences of murine

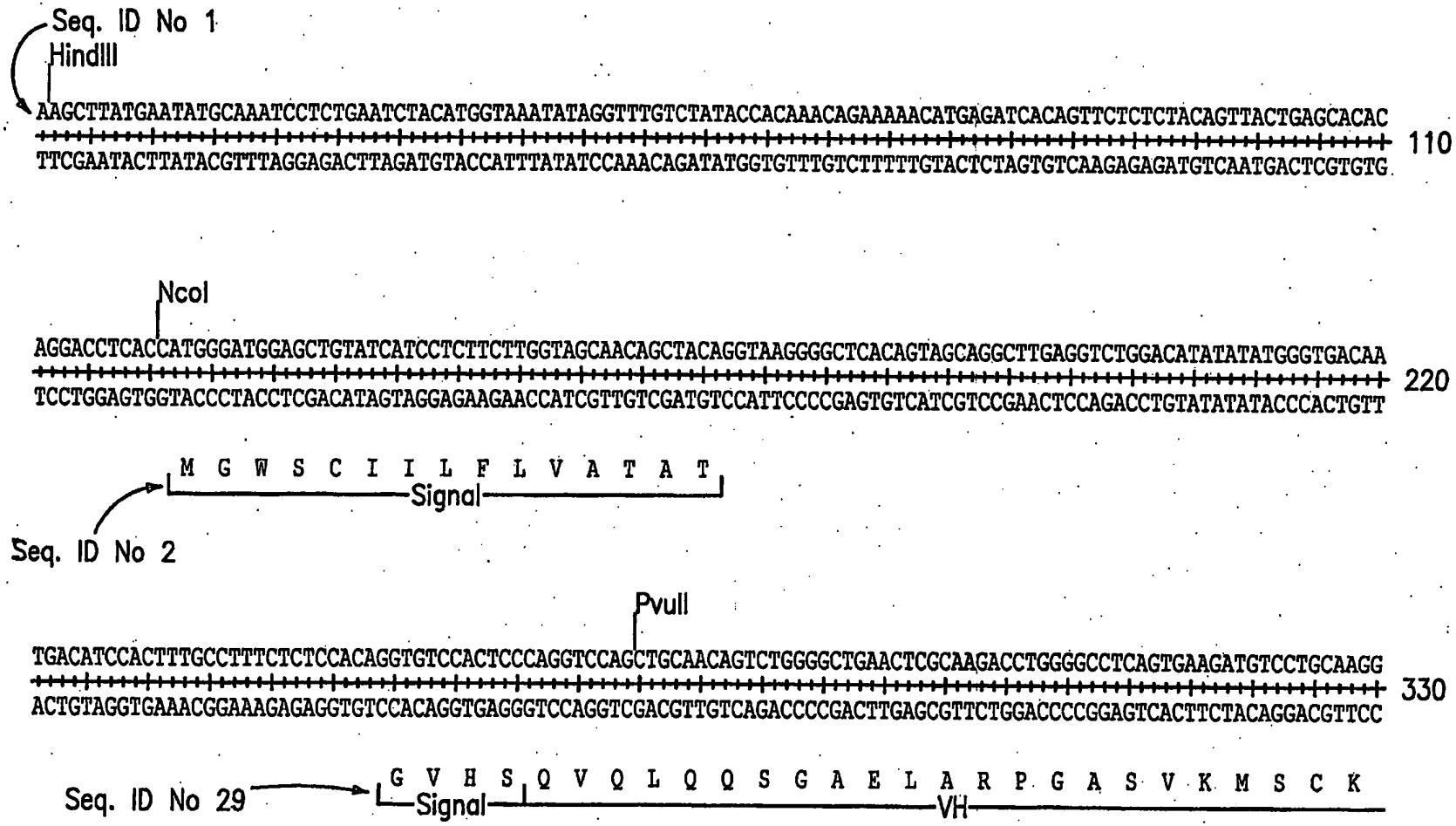


FIG. 3

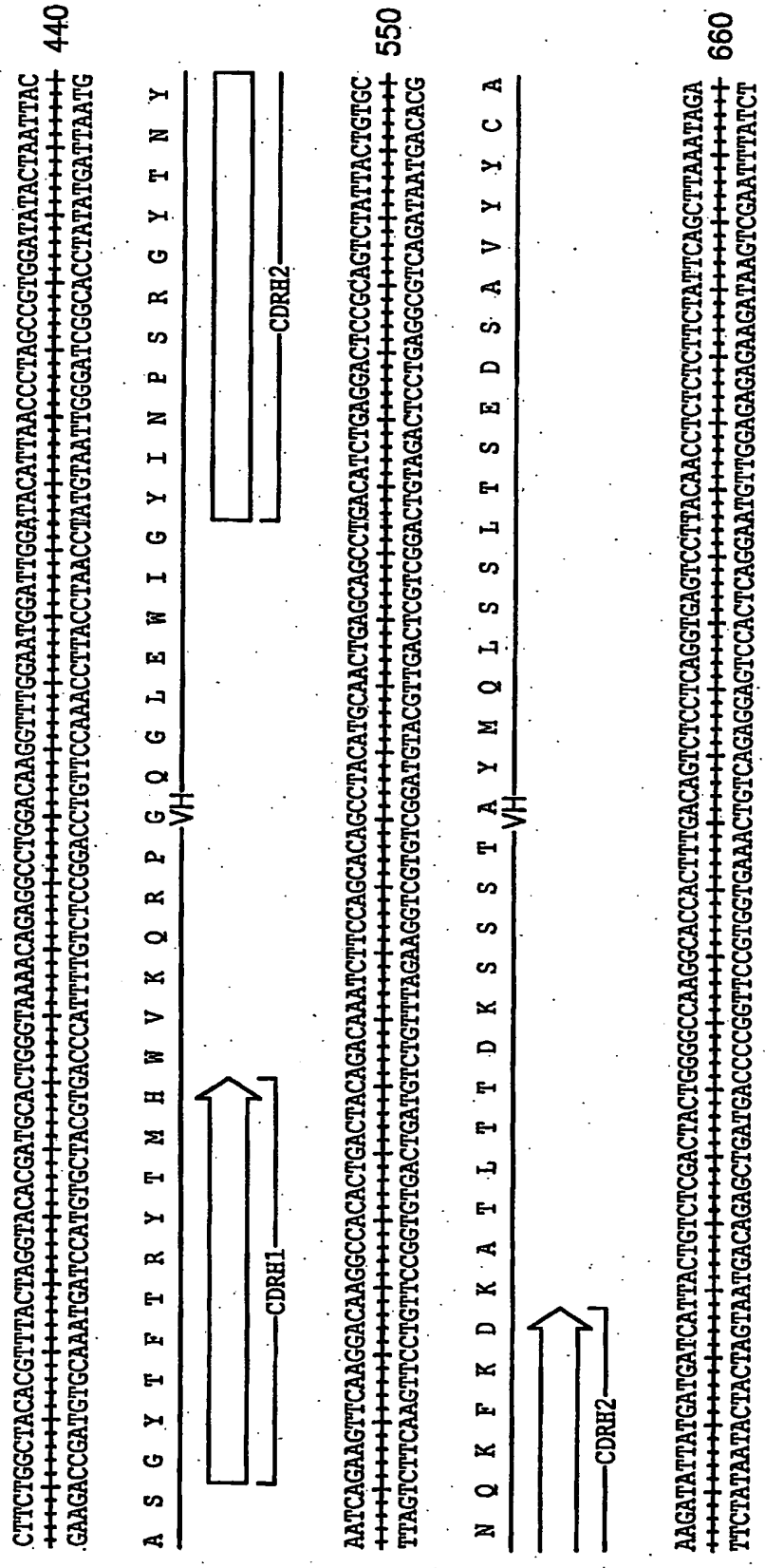
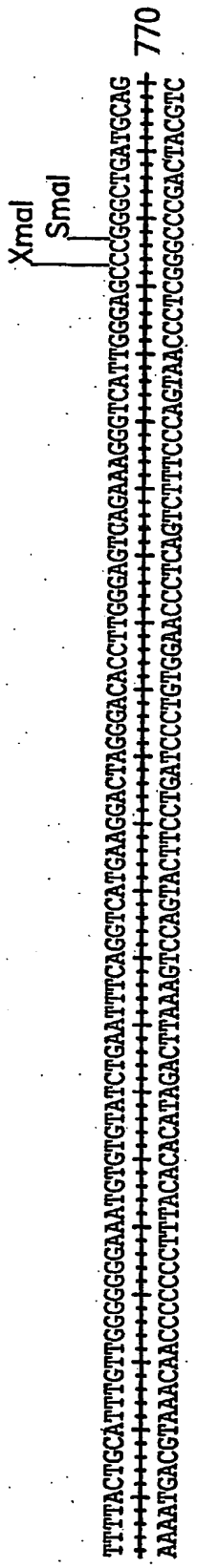


FIG. 3 (Cont.)


XmaI
SmaI
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AAAATGACGTAAACAACCCCCCCTTTACACACATAGACTTAAAGTCCAGTACTTCCTGATCCCTGTGGAACCCTCAGTCTTTCCCAGTAACCCTCGGGCCCGACTACGTC
770
BamHI
ACAGACATCCTCAGCTCCCAGACTTCATGGCCAGAGATTTATAGGATCC
TGTCTGTAGGAGTCGAGGGTCTGAAGTACCGGTCTCTAAATATCCTAGG
819

FIG. 3 (Cont.)

OKT3 VL gene construct. Nucleic acid and amino acid sequences of murine

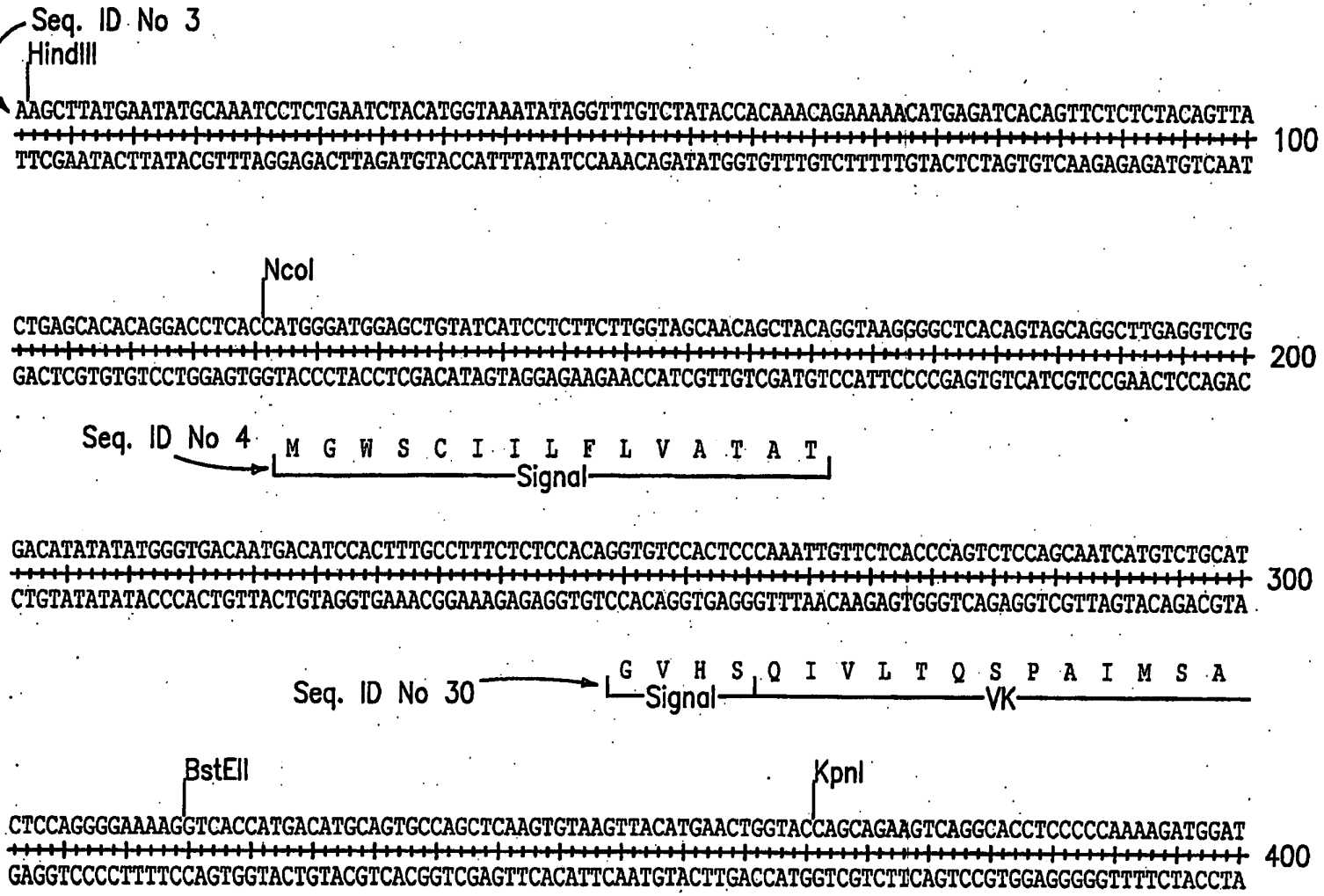
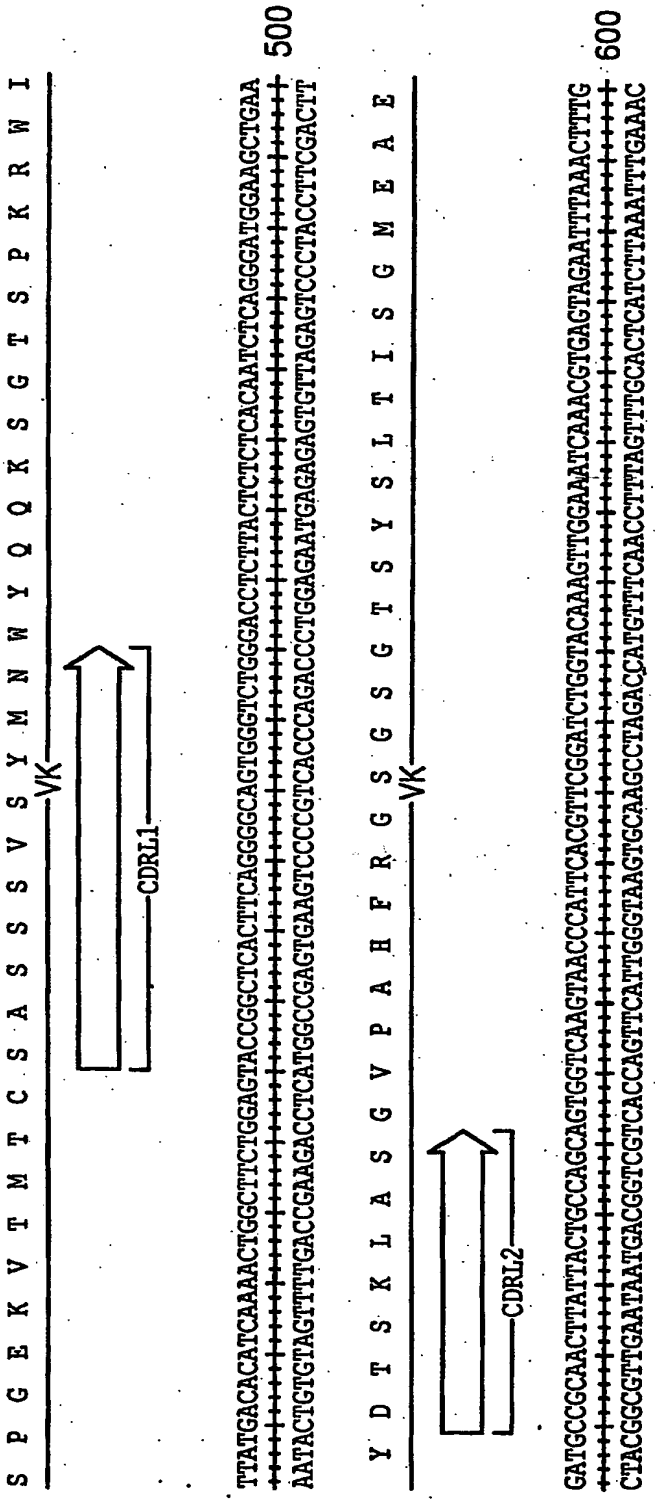


FIG. 4


S P G E K V T M T C S A S S S V S Y M N W Y Q Q K S G T S P K R W I
VK
CDRL1
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AATACTGTGTAGTTTTGACCGAAGACCTCATGGCCGAGTGAAGTCCCCGTCACCCAGACCCTGGAGAATGAGAGAGTGTTAGAGTCCCTACCTTCGACTT
500
Y D T S K L A S G V P A H F R G S G S G T S Y S L T I S G M E A E
VK
CDRL2
GATGCCGCAACTTATTACTGCCAGCAGTGGTCAAGTAACCCATTCACGTTCGGATCTGGTACAAAGTTGGAAATCAACGTGAGTAGAATTTAAACTTTG
CTACGGCGTTGAATAATGACGGTCGTCACCAGTTCATTGGGTAAGTGCAAGCCTAGACCATGTTTCAACCTTTAGTTTGCACTCATCTTAAATTTGAAAC
600
D A A T Y Y C Q Q W S S N P F T F G S G T K L E I K
VK
CDRL3
BamHI
CTTCCTCAGTTGGATCC
GAAGGAGTCAACCTAGG
617

FIG. 4 (Cont.)

Schematic map of the vector Apex−1 3F4V_HHuGamma4.

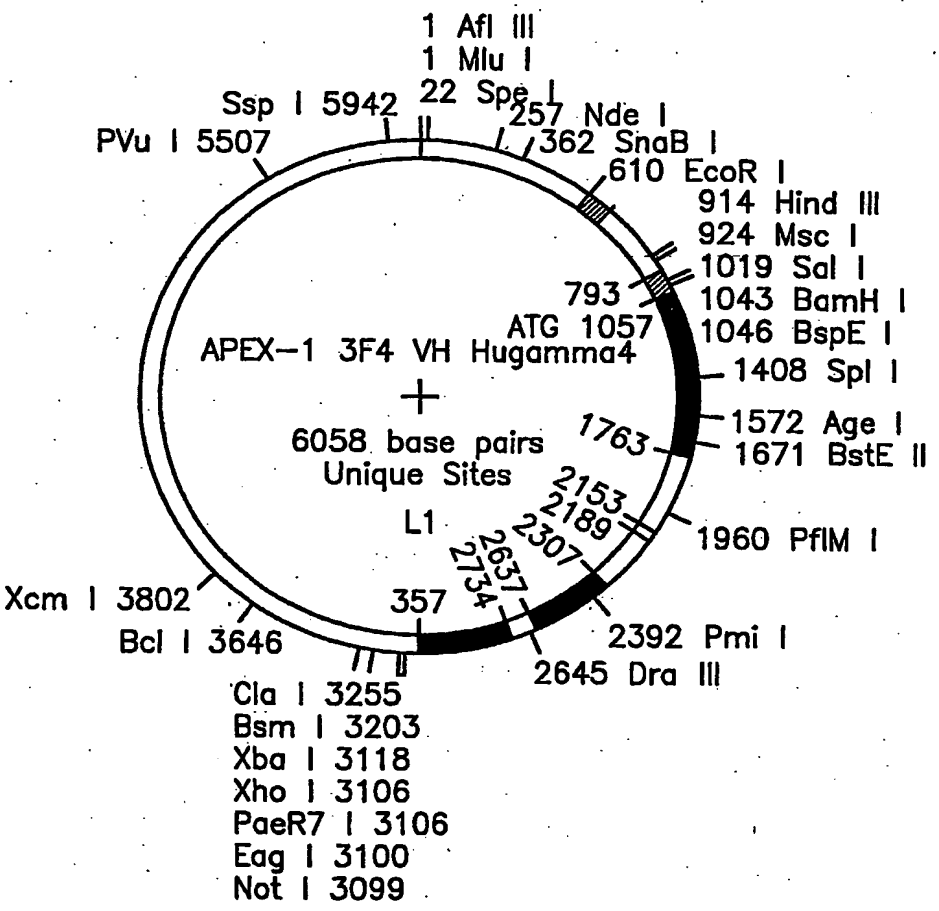


FIG. 5A

Vector sequence (APEX−1 3F4V$_H$HuGamma4)

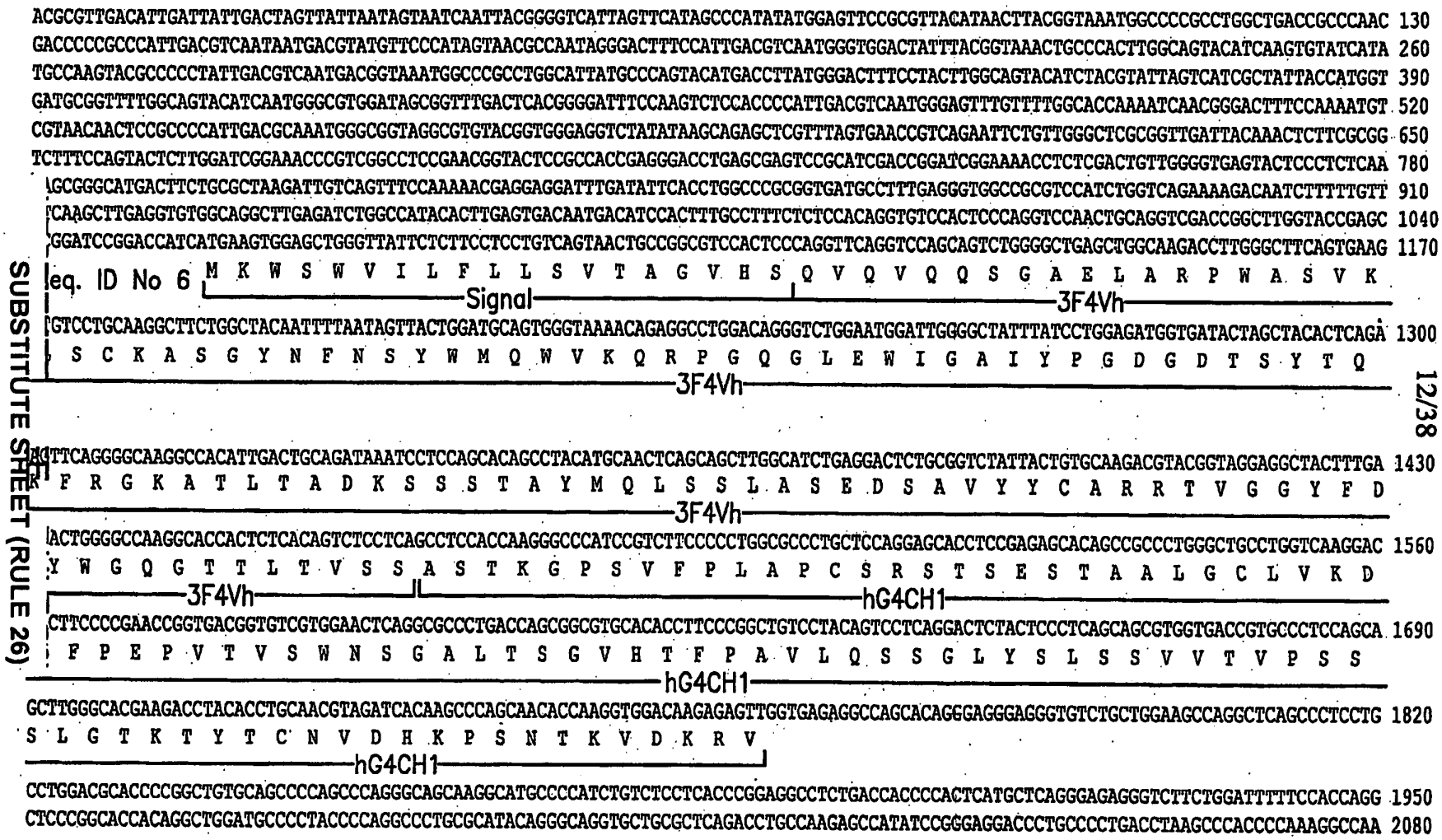
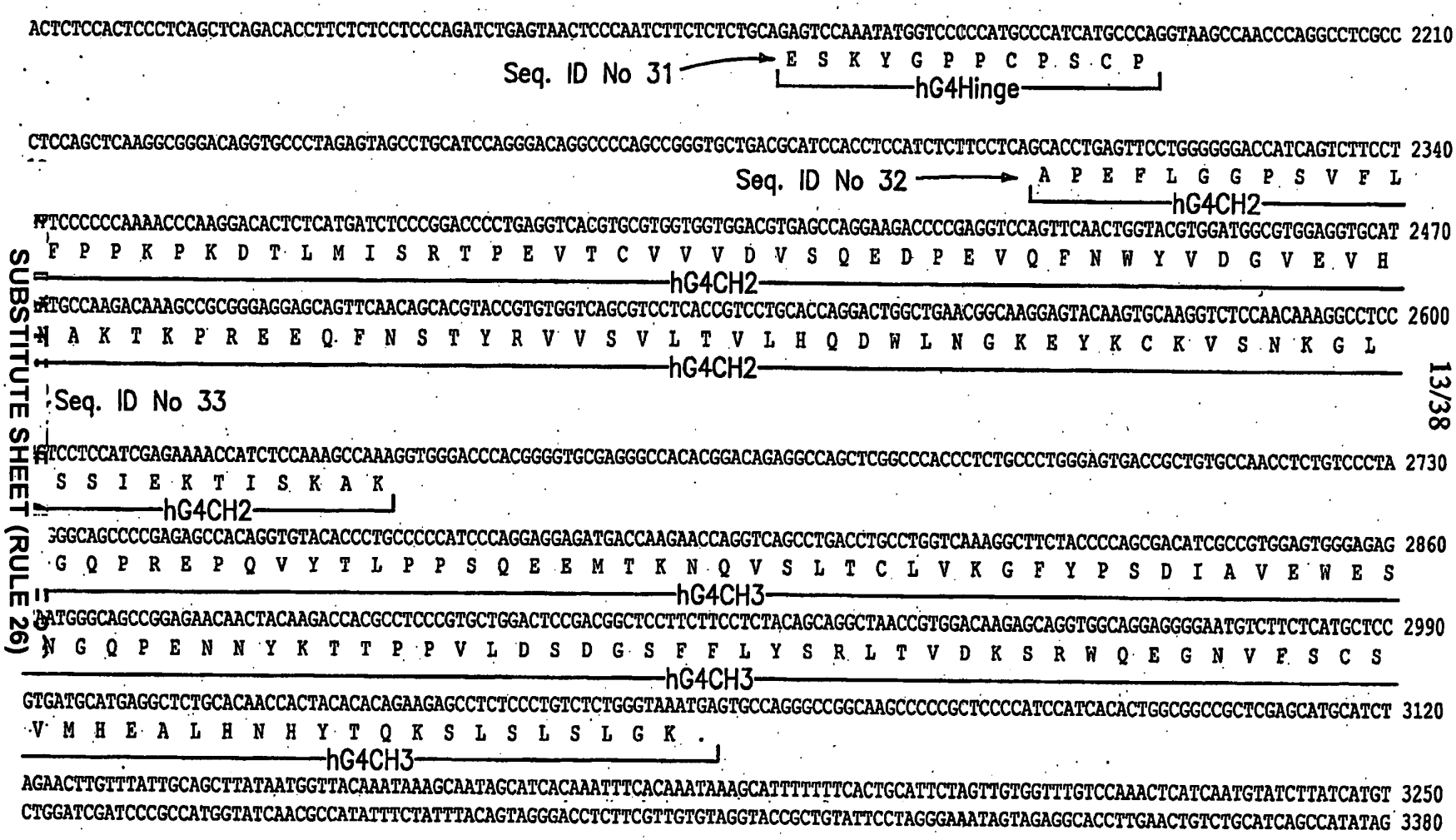


FIG. 5B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Seq. ID No 31
E S K Y G P P C P S C P
hG4Hinge
CTCCAGCTCAAGGCGGGACAGGTGCCCTAGAGTAGCCTGCATCCAGGGACAGGCCCCAGCCGGGTGCTGACGCATCCACCTCCATCTCTTCCTCAGCACCTGAGTTCCTGGGGGGACCATCAGTCTTCCT 2340
Seq. ID No 32
A P E F L G G P S V F L
hG4CH2
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F P P K P K D T L M I S R T P E V T C V V V D V S Q E D P E V Q F N W Y V D G V E V H
hG4CH2
ATGCCAAGACAAAGCCGCGGGAGGAGCAGTTCAACAGCACGTACCGTGTGGTCAGCGTCCTCACCGTCCTGCACCAGGACTGGCTGAACGGCAAGGAGTACAAGTGCAAGGTCTCCAACAAAGGCCTCC 2600
N A K T K P R E E Q F N S T Y R V V S V L T V L H Q D W L N G K E Y K C K V S N K G L
hG4CH2
Seq. ID No 33
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S S I E K T I S K A K
hG4CH2
GGGCAGCCCCGAGAGCCACAGGTGTACACCCTGCCCCCATCCCAGGAGGAGATGACCAAGAACCAGGTCAGCCTGACCTGCCTGGTCAAAGGCTTCTACCCCAGCGACATCGCCGTGGAGTGGGAGAG 2860
G Q P R E P Q V Y T L P P S Q E E M T K N Q V S L T C L V K G F Y P S D I A V E W E S
hG4CH3
AATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGCTGGACTCCGACGGCTCCTTCTTCCTCTACAGCAGGCTAACCGTGGACAAGAGCAGGTGGCAGGAGGGGAATGTCTTCTCATGCTCC 2990
N G Q P E N N Y K T T P P V L D S D G S F F L Y S R L T V D K S R W Q E G N V F S C S
hG4CH3
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V M H E A L H N H Y T Q K S L S L S L G K .
hG4CH3
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CTGGATCGATCCCGCCATGGTATCAACGCCATATTTCTATTTACAGTAGGGACCTCTTCGTTGTGTAGGTACCGCTGTATTCCTAGGGAAATAGTAGAGGCACCTTGAACTGTCTGCATCAGCCATATAG 3380

FIG. 5B (Cont.)

GGCCCGCTGTTCGACTTACAAACACAGGCACAGTACTGACAAACCCATACACCTCCTCTGAAATACCCATAGTTGCTAGGGCTGTCTCCGAACTCATTACACCCTCCAAAGTCAGAGCTGTAATTTCGCC 3510
ATCAAGGGCAGCGAGGGCTTCTCCAGATAAAATAGCTTCTGCCGAGAGTCCCGTAAGGGTAGACACTTCAGCTAATCCCTCGATGAGGTCTACTAGAATAGTCAGTGCGGCTCCCATTTTGAAAATTCAC 3640
GTACTTGATCAGCTTCAGAAGATGGCGGAGGGCCTCCAACACAGTAATTTTCCTCCCGACTCTTAAAATAGAAAATGTCAAGTCAGTTAAGCAGGAAGTGGACTAACTGACGCAGCTGGCCGTGCGACAT 3770
CCTCTTTTAATTAGTTGCTAGGCAACGCCCTCCAGAGGGCGTGTGGTTTTGCAAGAGGAAGCAAAAGCCTCTCCACCCAGGCCTAGAATGTTTCCACCCAATCATTACTATGACAACAGCTGTTTTTTTT 3900
AGTATTAAGCAGAGGCCGGGGACCCCTGGGCCCGCTTACTCTGGAGAAAAAGAAGAGAGGCATTGTAGAGGCTTCCAGAGGCAACTTGTCAAAACAGGACTGCTTCTATTTCTGTCACACTGTCTGGCCC 4030
TGTCACAAGGTCCAGCACCTCCATACCCCCTTTAATAAGCAGTTTGGGAACGGGTGCGGGTCTTACTCCGCCCATCCCGCCCCTAACTCCGCCCAGTTCCGCCCATTCTCCGCCCCATGGCTGACTAATT 4160
TTTTTTATTTATGCAGAGGCCGAGGCCGCCTCGGCCTCTGAGCTATTCCAGAAGTAGTGAGGAGGCTTTTTTGGAGGCCTAGGCTTTTGCAAAAAGGAGCTCCCAGCAAAAGGCCAGGAACCGTAAAAAG 4290
GCCGCGTTGCTGGCGTTTTTCCATAGGCTCCGCCCCCCTGACGAGCATCACAAAAATCGACGCTCAAGTCAGAGGTGGCGAAACCCGACAGGACTATAAAGATACCAGGCGTTTCCCCCTGGAAGCTCCC 4420
TCGTGCGCTCTCCTGTTCCGACCCTGCCGCTTACCGGATACCTGTCCGCCTTTCTCCCTTCGGGAAGCGTGGCGCTTTCTCAATGCTCACGCTGTAGGTATCTCAGTTCGGTGTAGGTCGTTCGCTCCAA 4550
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CTTGATCCGGCAAACAAACCACCGCTGGTAGCGGTGGTTTTTTTGTTTGCAAGCAGCAGATTACGCGCAGAAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTTCTACGGGGTCTGACGCTCAGTGGAA 4940
CGAAAACTCACGTTAAGGGATTTTGGTCATGAGATTATCAAAAAGGATCTTCACCTAGATCCTTTTAAATTAAAAATGAAGTTTTAAATCAATCTAAAGTATATATGAGTAAACTTGGTCTGACAGTTAC 5070
CAATGCTTAATCAGTGAGGCACCTATCTCAGCGATCTGTCTATTTCGTTCATCCATAGTTGCCTGACTCCCCGTCGTGTAGATAACTACGATACGGGAGGGCTTACCATCTGGCCCCAGTGCTGCAATGA 5200
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TGATCCCCCATGTTGTGCAAAAAAGCGGTTAGCTCCTTCGGTCCTCCGATCGTTGTCAGAAGTAAGTTGGCCGCAGTGTTATCACTCATGGTTATGGCAGCACTGCATAATTCTCTTACTGTCATGCCAT 5590
CCGTAAGATGCTTTTCTGTGACTGGTGAGTACTCAACCAAGTCATTCTGAGAATAGTGTATGCGGCGACCGAGTTGCTCTTGCCCGGCGTCAATACGGGATAATACCGCGCCACATAGCAGAACTTTAAA 5720
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GCGGATACATATTTGAATGTATTTAGAAAAATAAACAAATAGGGGTTCCGCGCACATTTCCCCGAAAAGTGCCACCTG 6058

FIG. 5B (Cont.)

SUBSTITUTE SHEET (RULE 26)

Schematic map of the vector Apex−1 3F4V_HHuG2/G4.

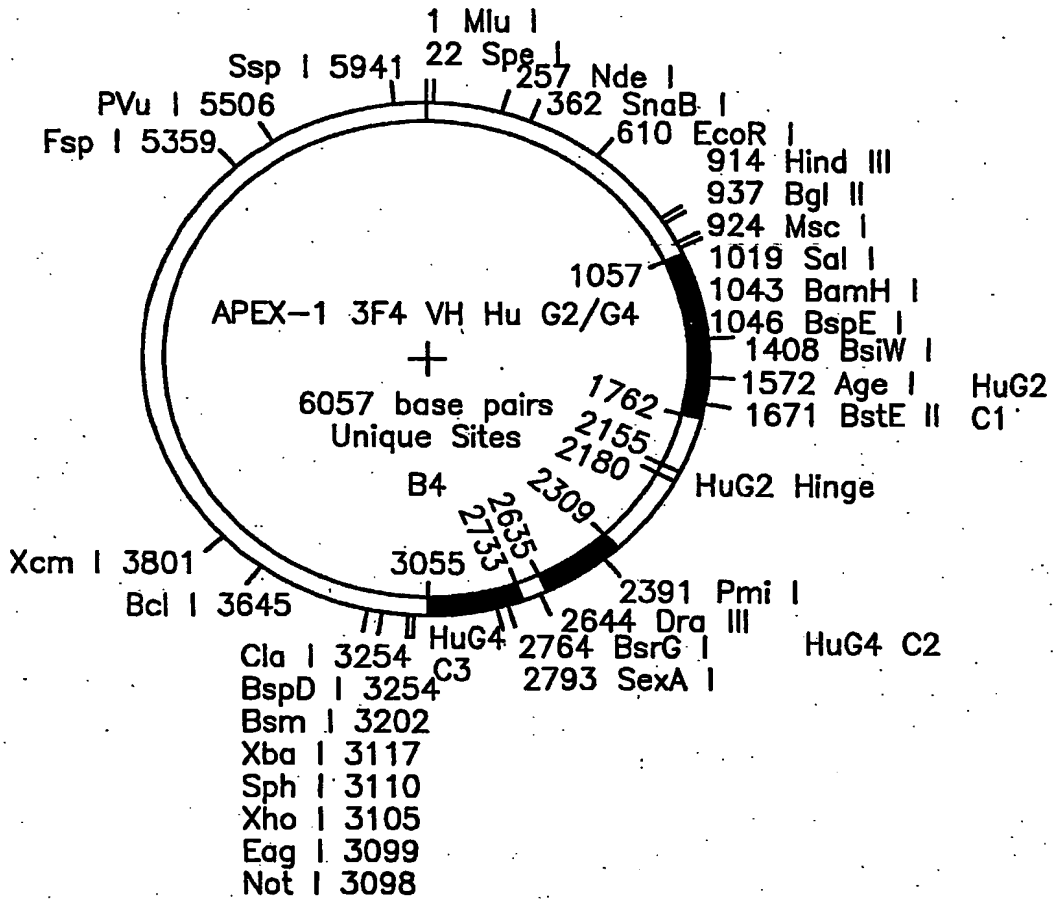

FIG. 6A

Vector Sequence (APEX−13F4V$_H$HuG2/G4)

```
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AGTACATCAAGTGTATCATATGCCAAGTACGCCCCCTATTGACGTCAATGACGGTAAATGGCCCGCCTGGCATTATGCCCAGTACATGACCTTATGGGACTTTCCTACTTGGCAGTACAT 360
CTACGTATTAGTCATCGCTATTACCATGGTGATGCGGTTTTGGCAGTACATCAATGGGCGTGGATAGCGGTTTGACTCACGGGGATTTCCAAGTCTCCACCCCATTGACGTCAATGGGAG 480
TTTGTTTTGGCACCAAAATCAACGGGACTTTCCAAAATGTCGTAACAACTCCGCCCCATTGACGCAAATGGGCGGTAGGCGTGTACGGTGGGAGGTCTATATAAGCAGAGCTCGTTTAGT 600
AACCGTCAGAATTCTGTTGGGCTCGCGGTTGATTACAAACTCTTCGCGGTCTTTCCAGTACTCTTGGATCGGAAACCCGTCGGCCTCCGAACGGTACTCCGCCACCGAGGGACCTGAGC 720
AGTCCGCATCGACCGGATCGGAAAACCTCTCGACTGTTGGGGTGAGTACTCCCTCTCAAAAGCGGGCATGACTTCTGCGCTAAGATTGTCAGTTTCCAAAAACGAGGAGGATTTGATAT 840
CACCTGGCCCGCGGTGATGCCTTTGAGGGTGGCCGCGTCCATCTGGTCAGAAAAGACAATCTTTTTGTTGTCAAGCTTGAGGTGTGGCAGGCTTGAGATCTGGCCATACACTTGAGTGA 960
AATGACATCCACTTTGCCTTTCTCTCCACAGGTGTCCACTCCCAGGTCCAACTGCAGGTCGACCGGCTTGGTACCGAGCTCGGATCCGGACCATCATGAAGTGGAGCTGGGTTATTCTC 1080
                                                                    (Seq. ID No: 8) → M K W S W V I L
                                                                                    └ Signal ┘
TCCTCCTGTCAGTAACTGCCGGCGTCCACTCCCAGGTTCAGGTCCAGCAGTCTGGGGCTGAGCTGGCAAGACCTTGGGCTTCAGTGAAGTTGTCCTGCAAGGCTTCTGGCTACAATTTT 1200
? L L S V T A G V H S Q V Q V Q Q S G A E L A R P W A S V K L S C K A S G Y N F
└ Signal ┘                                  └ 3F4Vh ┘
ATAGTTACTGGATGCAGTGGGTAAAACAGAGGCCTGGACAGGGTCTGGAATGGATTGGGGCTATTTATCCTGGAGATGGTGATACTAGCTACACTCAGAAGTTCAGGGGCAAGGCCACA 1320
N S Y W M Q W V K Q R P G Q G L E W I G A I Y P G D G D T S Y T Q K F R G K A T
                                  └ 3F4Vh ┘
TGACTGCAGATAAATCCTCCAGCACAGCCTACATGCAACTCAGCAGCTTGGCATCTGAGGACTCTGCGGTCTATTACTGTGCAAGACGTACGGTAGGAGGCTACTTTGACTACTGGGGC 1440
L T A D K S S S T A Y M Q L S S L A S E D S A V Y Y C A R R T V G G Y F D Y W G
                                  └ 3F4Vh ┘
CAAGGCACCACTCTCACAGTCTCCTCAGCCTCCACCAAGGGCCCATCCGTCTTCCCCCTGGCGCCCTGCTCCAGGAGCACCTCCGAGAGCACAGCCGCCCTGGGCTGCCTGGTCAAGGAC 1560
Q G T T L T V S S A S T K G P S V F P L A P C S R S T S E S T A A L G C L V K D
└ 3F4Vh ┘                                  └ G2G4CH1 ┘
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FIG. 6B

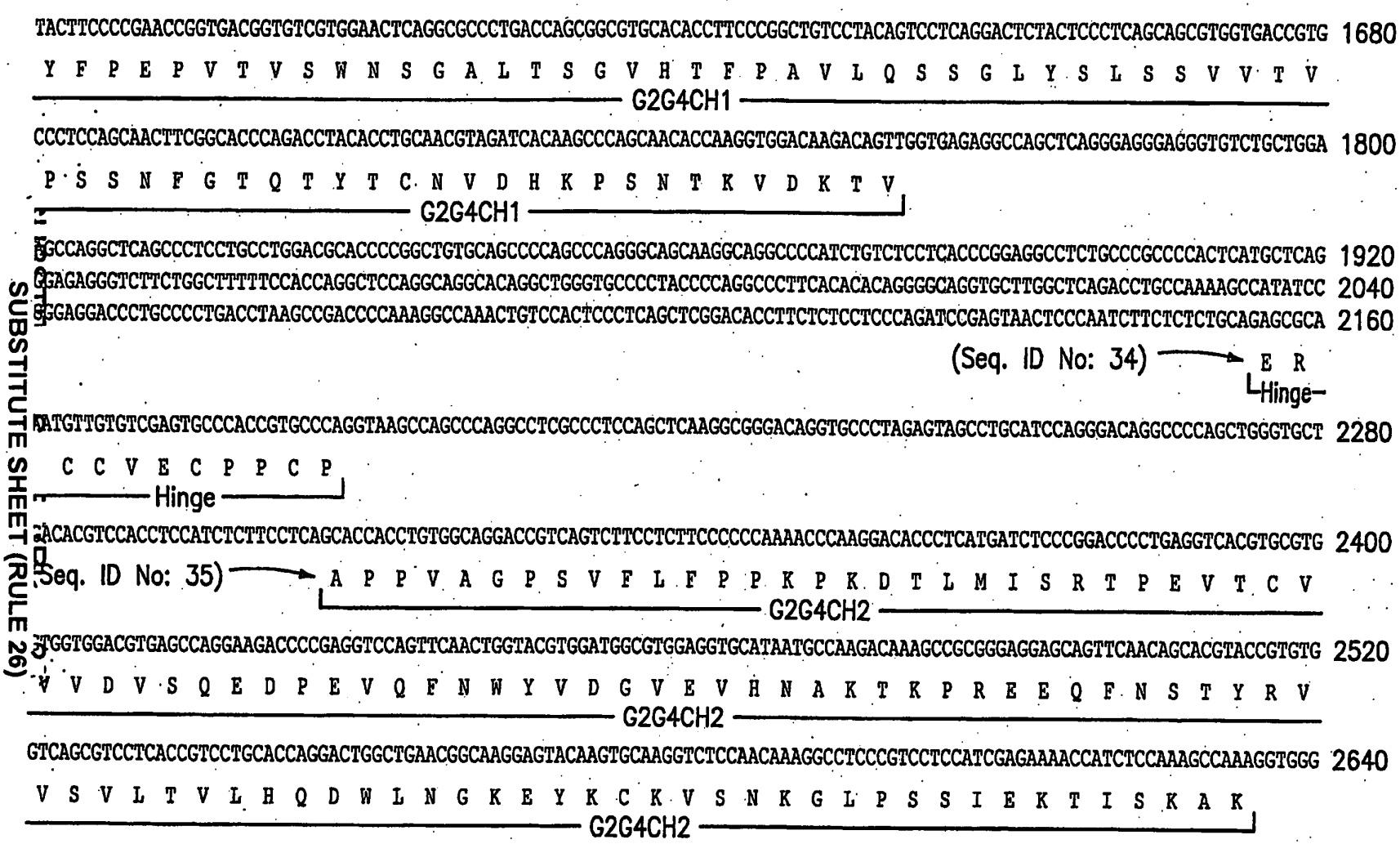

FIG. 6B (Cont.)

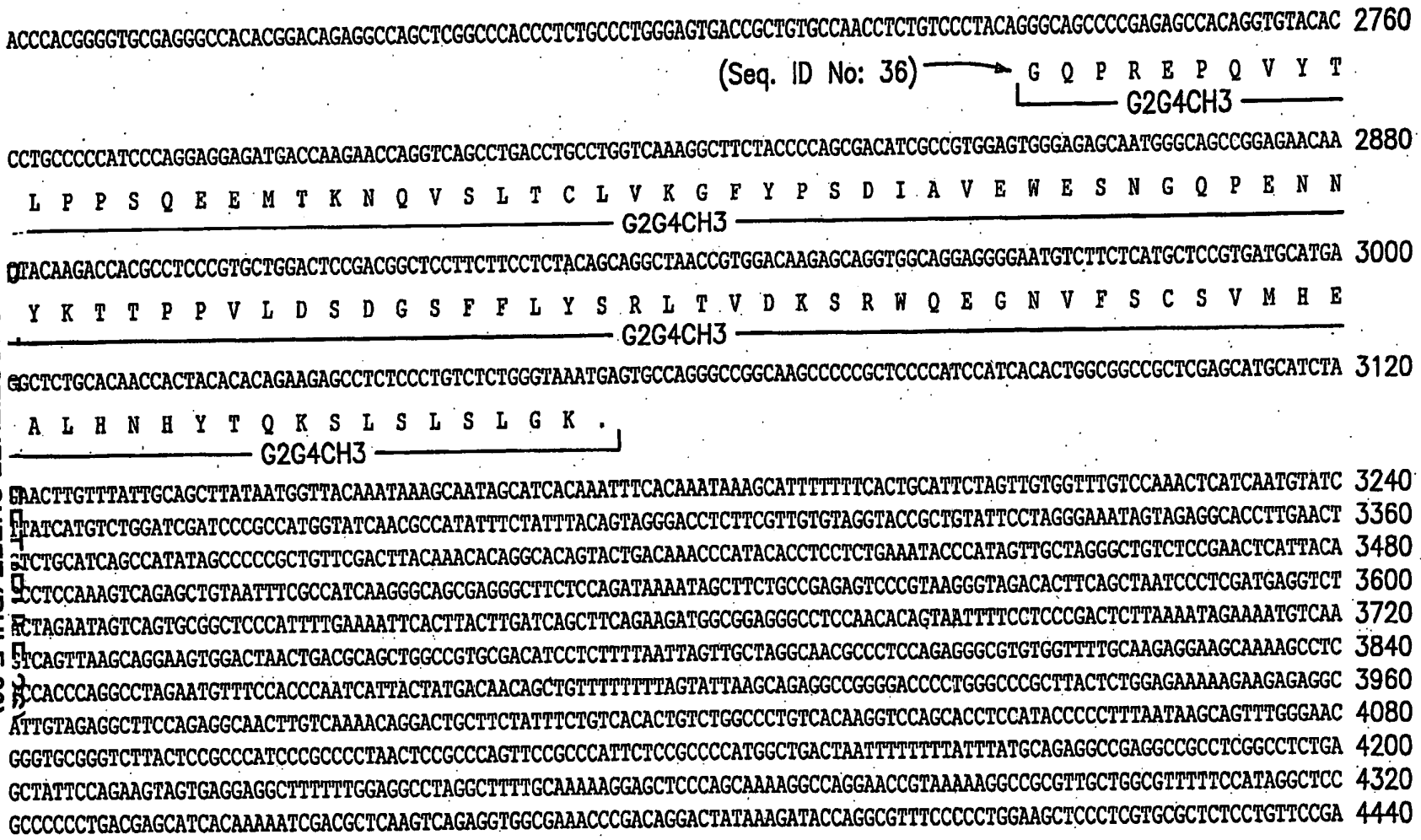

FIG. 6B (Cont.)

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GAGCGAGGTATGTAGGCGGTGCTACAGAGTTCTTGAAGTGGTGGCCTAACTACGGCTACACTAGAAGGACAGTATTTGGTATCTGCGCTCTGCTGAAGCCAGTTACCTTCGGAAAAAGAG 4800
TTGGTAGCTCTTGATCCGGCAAACAAACCACCGCTGGTAGCGGTGGTTTTTTTGTTTGCAAGCAGCAGATTACGCGCAGAAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTTCTACGG 4920
GGTCTGACGCTCAGTGGAACGAAAACTCACGTTAAGGGATTTTGGTCATGAGATTATCAAAAAGGATCTTCACCTAGATCCTTTTAAATTAAAAATGAAGTTTTAAATCAATCTAAAGTA 5040
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TACGGGAGGGCTTACCATCTGGCCCCAGTGCTGCAATGATACCGCGAGACCCACGCTCACCGGCTCCAGATTTATCAGCAATAAACCAGCCAGCCGGAAGGGCCGAGCGCAGAAGTGGTC 5280
CTGCAACTTTATCCGCCTCCATCCAGTCTATTAATTGTTGCCGGGAAGCTAGAGTAAGTAGTTCGCCAGTTAATAGTTTGCGCAACGTTGTTGCCATTGCTACAGGCATCGTGGTGTCAC 5400
GCTCGTCGTTTGGTATGGCTTCATTCAGCTCCGGTTCCCAACGATCAAGGCGAGTTACATGATCCCCCATGTTGTGCAAAAAAGCGGTTAGCTCCTTCGGTCCTCCGATCGTTGTCAGAA 5520
GTAAGTTGGCCGCAGTGTTATCACTCATGGTTATGGCAGCACTGCATAATTCTCTTACTGTCATGCCATCCGTAAGATGCTTTTCTGTGACTGGTGAGTACTCAACCAAGTCATTCTGAG 5640
AATAGTGTATGCGGCGACCGAGTTGCTCTTGCCCGGCGTCAATACGGGATAATACCGCGCCACATAGCAGAACTTTAAAAGTGCTCATCATTGGAAAACGTTCTTCGGGGCGAAAACTCT 5760
CAAGGATCTTACCGCTGTTGAGATCCAGTTCGATGTAACCCACTCGTGCACCCAACTGATCTTCAGCATCTTTTACTTTCACCAGCGTTTCTGGGTGAGCAAAAACAGGAAGGCAAAATG 5880
CCGCAAAAAAGGGAATAAGGGCGACACGGAAATGTTGAATACTCATACTCTTCCTTTTTCAATATTATTGAAGCATTTATCAGGGTTATTGTCTCATGAGCGGATACATATTTGAATGTA 6000
TTTAGAAAAATAAACAAATAGGGGTTCCGCGCACATTTCCCCGAAAAGTGCCACCTG 6057

FIG. 6B (Cont.)

Map of the heavy chain expression vector pSVgptHuG2/G4 used in

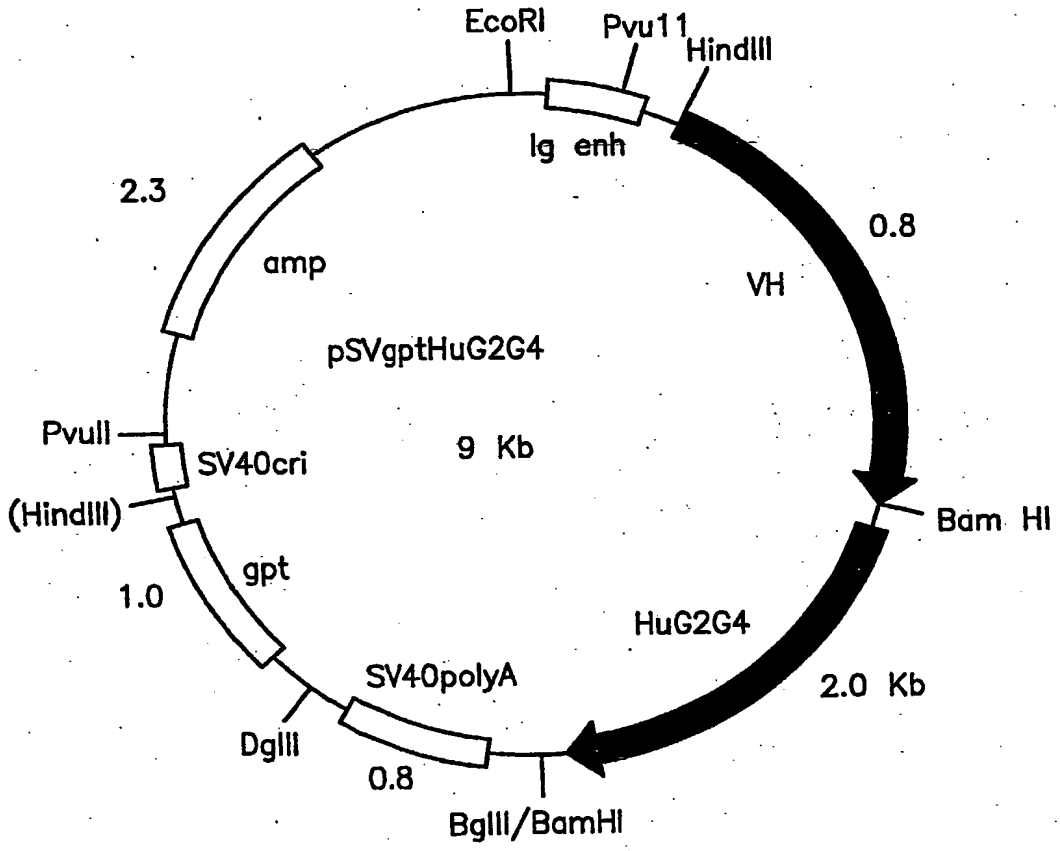


FIG. 7

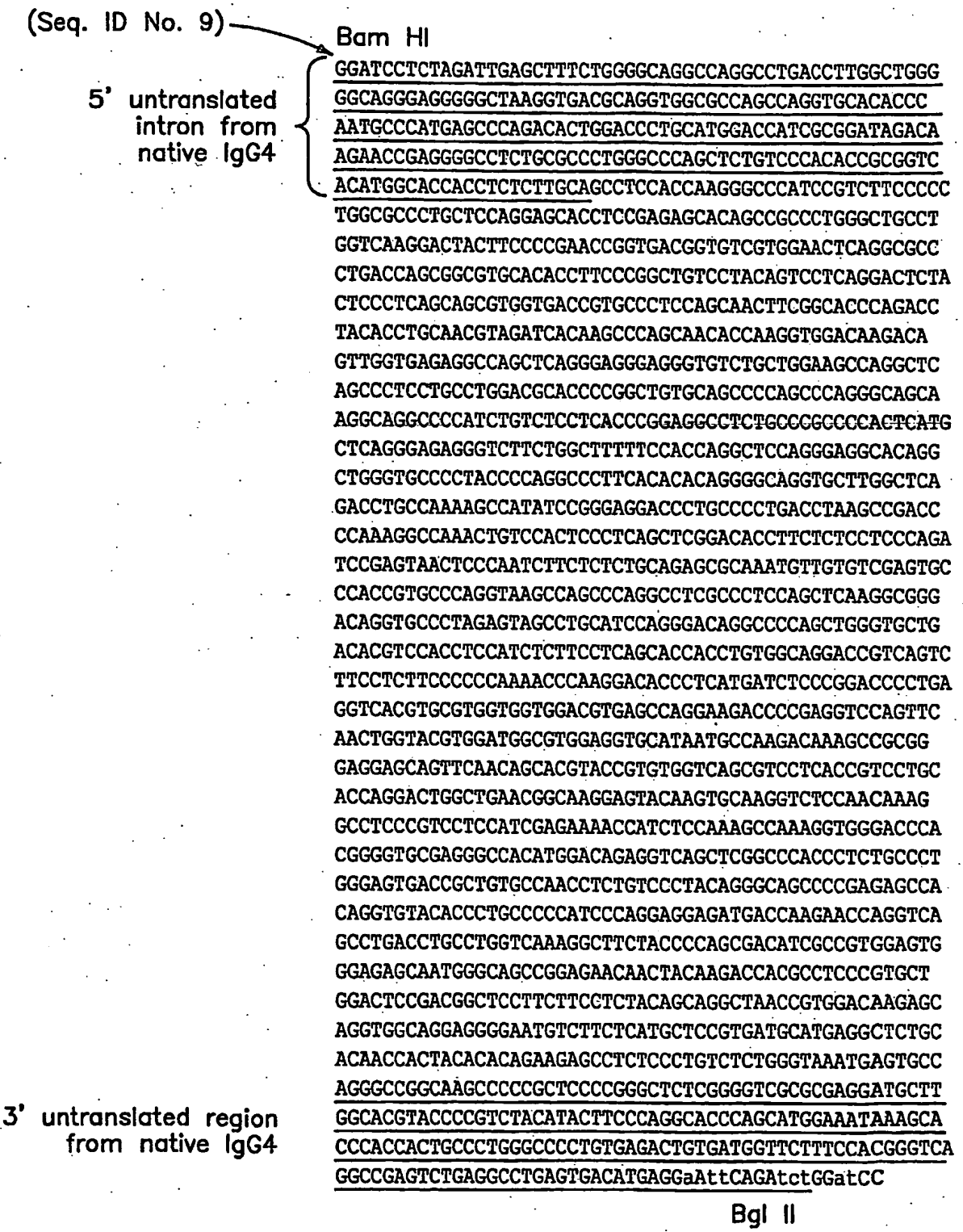

(Seq. ID No. 9)
Bam HI
5' untranslated intron from native IgG4
GGATCCTCTAGATTGAGCTTTCTGGGGCAGGCCAGGCCTGACCTTGGCTGGG
GGCAGGGAGGGGGCTAAGGTGACGCAGGTGGCGCCAGCCAGGTGCACACCC
AATGCCCATGAGCCCAGACACTGGACCCTGCATGGACCATCGCGGATAGACA
AGAACCGAGGGGCCTCTGCGCCCTGGGCCCAGCTCTGTCCCACACCGCGGTC
ACATGGCACCACCTCTCTTGCAGCCTCCACCAAGGGCCCATCCGTCTTCCCCC
TGGCGCCCTGCTCCAGGAGCACCTCCGAGAGCACAGCCGCCCTGGGCTGCCT
GGTCAAGGACTACTTCCCCGAACCGGTGACGGTGTCGTGGAACTCAGGCGCC
CTGACCAGCGGCGTGCACACCTTCCCGGCTGTCCTACAGTCCTCAGGACTCTA
CTCCCTCAGCAGCGTGGTGACCGTGCCCTCCAGCAACTTCGGCACCCAGACC
TACACCTGCAACGTAGATCACAAGCCCAGCAACACCAAGGTGGACAAGACA
GTTGGTGAGAGGCCAGCTCAGGGAGGGAGGGTGTCTGCTGGAAGCCAGGCTC
AGCCCTCCTGCCTGGACGCACCCCGGCTGTGCAGCCCCAGCCCAGGGCAGCA
AGGCAGGCCCCATCTGTCTCCTCACCCGGAGGCCTCTGCCCGCCCCACTCATG
CTCAGGGAGAGGGTCTTCTGGCTTTTTCCACCAGGCTCCAGGGAGGCACAGG
CTGGGTGCCCCTACCCCAGGCCCTTCACACACAGGGGCAGGTGCTTGGCTCA
GACCTGCCAAAAGCCATATCCGGGAGGACCCTGCCCCTGACCTAAGCCGACC
CCAAAGGCCAAACTGTCCACTCCCTCAGCTCGGACACCTTCTCTCCTCCCAGA
TCCGAGTAACTCCCAATCTTCTCTCTGCAGAGCGCAAATGTTGTGTCGAGTGC
CCACCGTGCCCAGGTAAGCCAGCCCAGGCCTCGCCCTCCAGCTCAAGGCGGG
ACAGGTGCCCTAGAGTAGCCTGCATCCAGGGACAGGCCCCAGCTGGGTGCTG
ACACGTCCACCTCCATCTCTTCCTCAGCACCACCTGTGGCAGGACCGTCAGTC
TTCCTCTTCCCCCCAAAACCCAAGGACACCCTCATGATCTCCCGGACCCCTGA
GGTCACGTGCGTGGTGGTGGACGTGAGCCAGGAAGACCCCGAGGTCCAGTTC
AACTGGTACGTGGATGGCGTGGAGGTGCATAATGCCAAGACAAAGCCGCGG
GAGGAGCAGTTCAACAGCACGTACCGTGTGGTCAGCGTCCTCACCGTCCTGC
ACCAGGACTGGCTGAACGGCAAGGAGTACAAGTGCAAGGTCTCCAACAAAG
GCCTCCCGTCCTCCATCGAGAAAACCATCTCCAAAGCCAAAGGTGGGACCCA
CGGGGTGCGAGGGCCACATGGACAGAGGTCAGCTCGGCCCACCCTCTGCCCT
GGGAGTGACCGCTGTGCCAACCTCTGTCCCTACAGGGCAGCCCCGAGAGCCA
CAGGTGTACACCCTGCCCCCATCCCAGGAGGAGATGACCAAGAACCAGGTCA
GCCTGACCTGCCTGGTCAAAGGCTTCTACCCCAGCGACATCGCCGTGGAGTG
GGAGAGCAATGGGCAGCCGGAGAACAACTACAAGACCACGCCTCCCGTGCT
GGACTCCGACGGCTCCTTCTTCCTCTACAGCAGGCTAACCGTGGACAAGAGC
AGGTGGCAGGAGGGGAATGTCTTCTCATGCTCCGTGATGCATGAGGCTCTGC
ACAACCACTACACACAGAAGAGCCTCTCCCTGTCTCTGGGTAAATGAGTGCC
AGGGCCGGCAAGCCCCCGCTCCCCGGGCTCTCGGGGTCGCGCGAGGATGCTT
3' untranslated region from native IgG4
GGCACGTACCCCGTCTACATACTTCCCAGGCACCCAGCATGGAAATAAAGCA
CCCACCACTGCCCTGGGCCCCTGTGAGACTGTGATGGTTCTTTCCACGGGTCA
GGCCGAGTCTGAGGCCTGAGTGACATGAGGaAttCAGAtctGGatCC
Bgl II

FIG. 8

Map of the light chain expression vector pSVgptHuCK

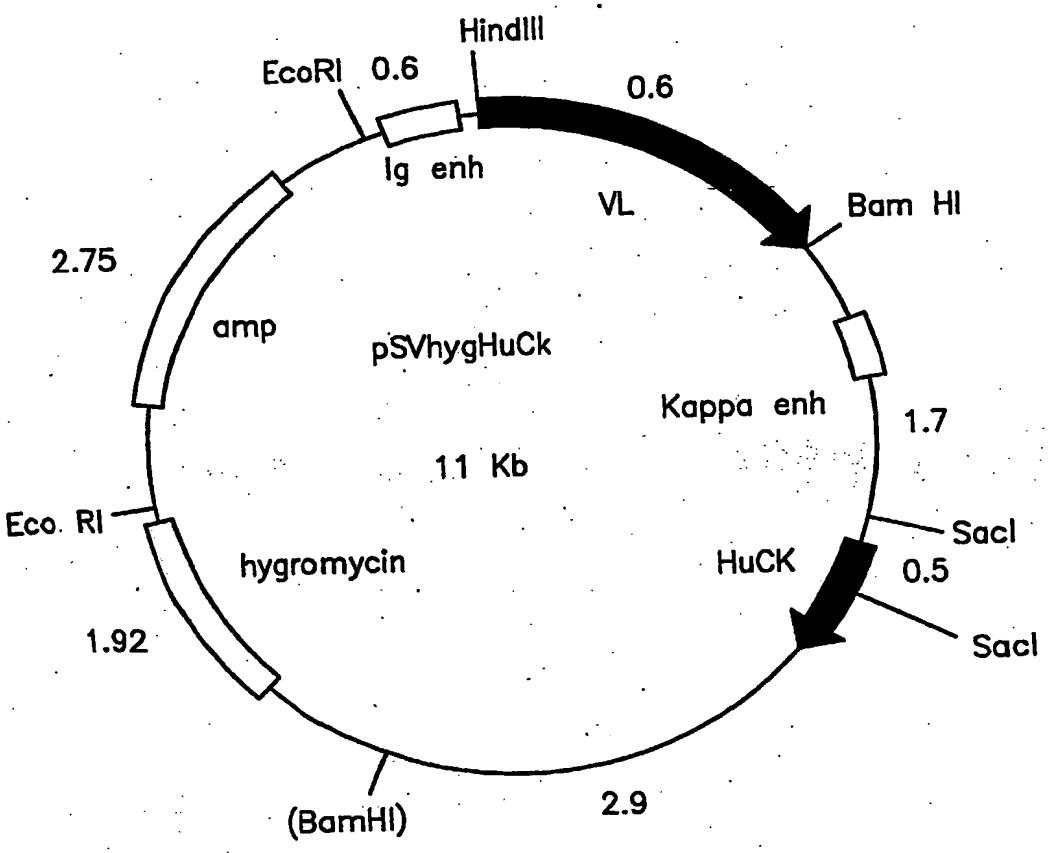


FIG. 9

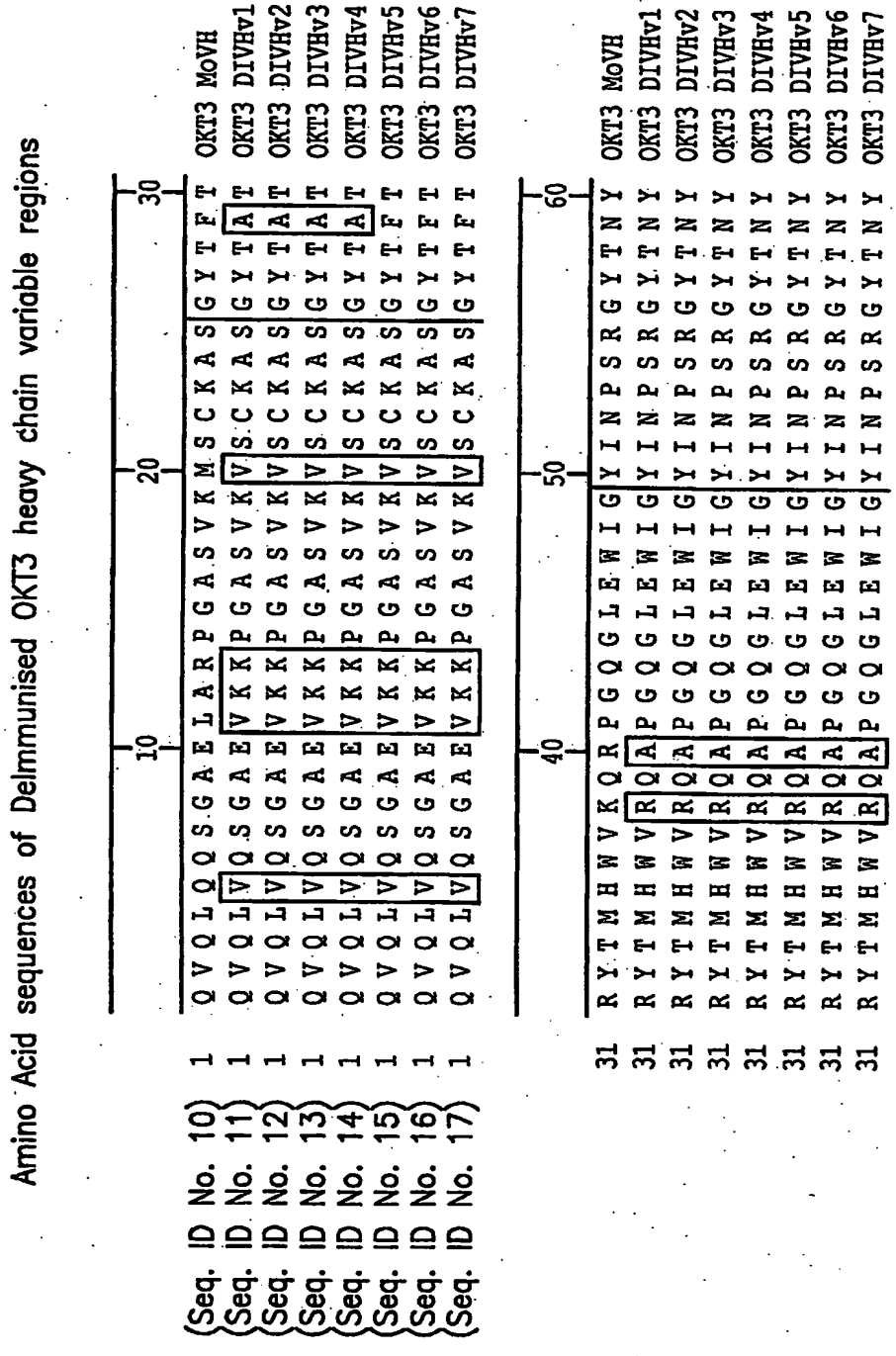

Amino Acid sequences of Delmmunised OKT3 heavy chain variable regions
(Seq. ID No. 10) 1 QVQLQQSGAELARPGASVKMSCKASGYTFT OKT3 MoVH
(Seq. ID No. 11) 1 QVQLVQSGAEVKKPGASVKVSCKASGYTAT OKT3 DIVHv1
(Seq. ID No. 12) 1 QVQLVQSGAEVKKPGASVKVSCKASGYTAT OKT3 DIVHv2
(Seq. ID No. 13) 1 QVQLVQSGAEVKKPGASVKVSCKASGYTAT OKT3 DIVHv3
(Seq. ID No. 14) 1 QVQLVQSGAEVKKPGASVKVSCKASGYTAT OKT3 DIVHv4
(Seq. ID No. 15) 1 QVQLVQSGAEVKKPGASVKVSCKASGYTFT OKT3 DIVHv5
(Seq. ID No. 16) 1 QVQLVQSGAEVKKPGASVKVSCKASGYTFT OKT3 DIVHv6
(Seq. ID No. 17) 1 QVQLVQSGAEVKKPGASVKVSCKASGYTFT OKT3 DIVHv7
10
20
30
40
50
60
31 RYTMHWVKQRPGQGLEWIGYINPSRGYTNY OKT3 MoVH
31 RYTMHWVRQAPGQGLEWIGYINPSRGYTNY OKT3 DIVHv1
31 RYTMHWVRQAPGQGLEWIGYINPSRGYTNY OKT3 DIVHv2
31 RYTMHWVRQAPGQGLEWIGYINPSRGYTNY OKT3 DIVHv3
31 RYTMHWVRQAPGQGLEWIGYINPSRGYTNY OKT3 DIVHv4
31 RYTMHWVRQAPGQGLEWIGYINPSRGYTNY OKT3 DIVHv5
31 RYTMHWVRQAPGQGLEWIGYINPSRGYTNY OKT3 DIVHv6
31 RYTMHWVRQAPGQGLEWIGYINPSRGYTNY OKT3 DIVHv7

FIG. 10

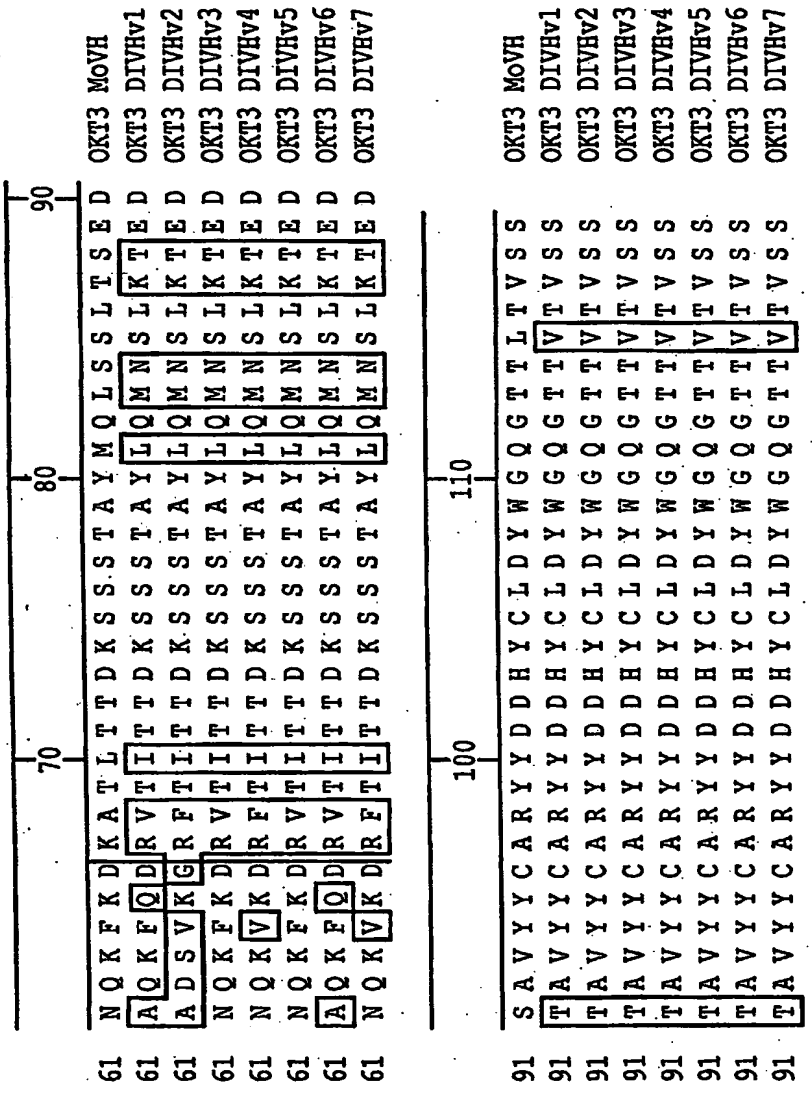

OKT3 MoVH
OKT3 DIVHv1
OKT3 DIVHv2
OKT3 DIVHv3
OKT3 DIVHv4
OKT3 DIVHv5
OKT3 DIVHv6
OKT3 DIVHv7
70
80
90
100
110
61
91

FIG. 10 (Cont.)

Amino Acid sequences of Delmmunised OKT3 light chain variable regions

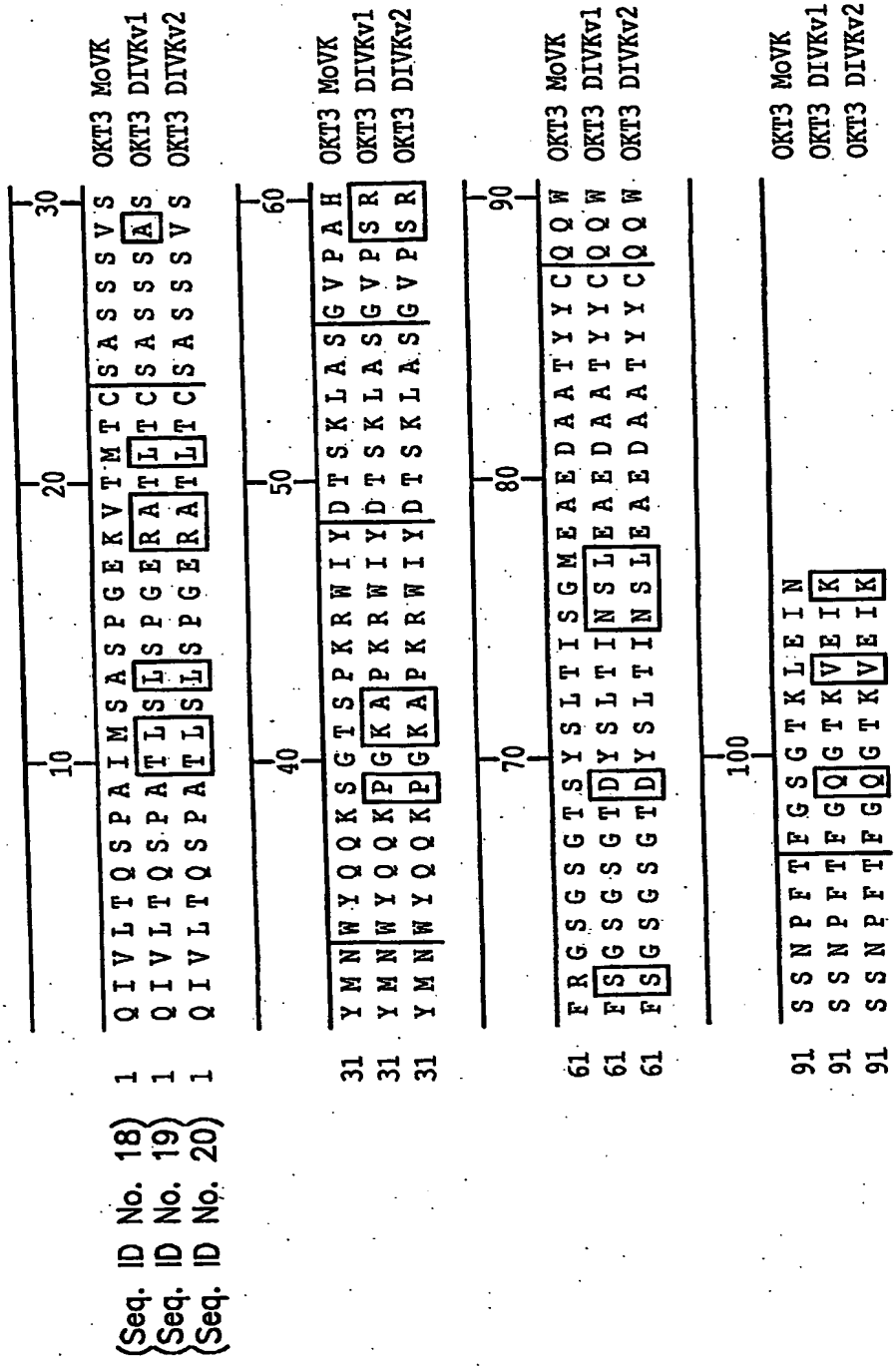


FIG. 11

OLIGOS FOR CONSTRUCTION OF DIVHs (SEQ ID NOS: 37–57)

KTDIVH1 GAAGTCAAGAAACCTGGGGCCTCAGTGAAGGTGTCCTGCAAGG
KTDIVH2 GCCCCAGGTTTCTTGACTTCAGCCCCAGACTGTACCAGCTGGACCTG
KTDIVH3 TGGGTAAGACAGGCGCCTGGACAAGGTTTGG
KTDIVH4 GTCCAGGCGCCTGTCTTACCCAGTGCATC
KTDIVH4A AGGCGCCTGTCTTACCCAGTGCATCGTGTACCTAGTAGCCGTGTAGCC
KTDIVH5 CAATCAGAAGTTCAAGGACAGGGTCACAATCACTACAGACAAA
KTDIVH5A CGCTCAGAAGTTCCAGGACAGGGTCACAATCACTACAGACAAA
KTDIVH5B CGCTGACAGTGTCAAGGGCAGGTTCACAATCACTACAGACAAA
KTDIVH5C CAATCAGAAGGTCAAGGACAGGTTCACAATCACTACAGACAAA
KTDIVH6 GTCCTTGAACTTCTGATTGTAATTAGTATATCCACGG
KTDIVH6A GTCCTGGAACTTCTGAGCGTAATTAGTATATCCACGG
KTDIVH6B GCCCTTGACACTGTCAGCGTAATTAGTATATCCACGG
KTDIVH6C GTCCTTGACCTTCTGATTGTAATTAGTATATCCACGG
KTDIVH7 AGCCTGAAAACTGAGGACACCGCAGTCTATTACTG
KTDIVH8 GTCCTCAGTTTTCAGGCTGTTCATTTGCAAGTAGGCTGTGCT
KTDIVH9 CCAAGGCACCACTGTGACAGTCTCCTCAGG
KTDIVH10 CCTGAGGAGACTGTCACAGTGGTGCCTTGG
KT3VHY GGTGTCCACTCCCAGGTCCAGCTG
KT3VHZ CAGCTGGACCTGGGAGTGGACACCTGTGG
VHVK1 GCATGTTGACCCTGACGCAAGCTTATGAATATGCAAA
VH12 GCGATAGCTGGACTGAATGGATCCTATAAATCTCTG

OLIGOS FOR CONSTRUCTION OF DIVKs (SEQ ID NOS: 58–74)

KTDIVK1 CCCTCTCTCTTTCTCCAGGGGAACGCGCCACCTTGACATGCAGTG
KTDIVK2 CCTGGAGAAAGAGAGAGGGTTGCTGGAGACTGGGTG
KTDIVK3 CATGAACTGGTACCAGCAGAAGCCCGGCAAAGCTCCCAAAAGATGGAT
KTDIVK4 CGGGCTTCTGCTGGTACCAGTTCATGTAACTTACACTT
KTDIVK4A CTTCTGCTGGTACCAGTTCATGTAACTTGCACTTGAGC
KTDIVK5 GGGTCTGGGACCGATTACTCTCTCACCATCAATAGTCTGGAAGCTGAAG
KTDIVK6 GTAATCGGTCCCAGACCCACTGCCACTGAAGCGAGACGGTACTCCAG
KTDIVK7 TTCACGTTCGGACAAGGTACAAAGGTGGAAATCAAACG
KTDIVK8 CTTTGTACCTTGTCCGAACGTGAATGGGTTACTTGACC
KKT22 GCGGATCCAGTCGACGAAGCA
KT3VKX CTGAATGGATCCAACTGAGGAAGCAAAGTTTAAATTCTACTCACG
KT3VKY CAAATTGTTCTCACCCAGTCTCCAGCAA
KT3VKZ TTGCTGGAGACTGGGTGAGAACAATTTGGGAG
KT3VKZ2 TGGAGACTGGGTGAGAACAATTTGGGAGTGGACACCTGTGG
KT3VKZ3 AGAGAGGGTTGCTGGAGACTGGGTGAGAACAATTTG
VHVK1 GCATGTTGACCCTGACGCAAGCTTATGAATATGCAAA
VK12 GCGATAGCTGGACTGAATGGATCCAACTGAGGAAGC

DNA and Amino acid sequence of Delmmunised OKT3 VH version 1.

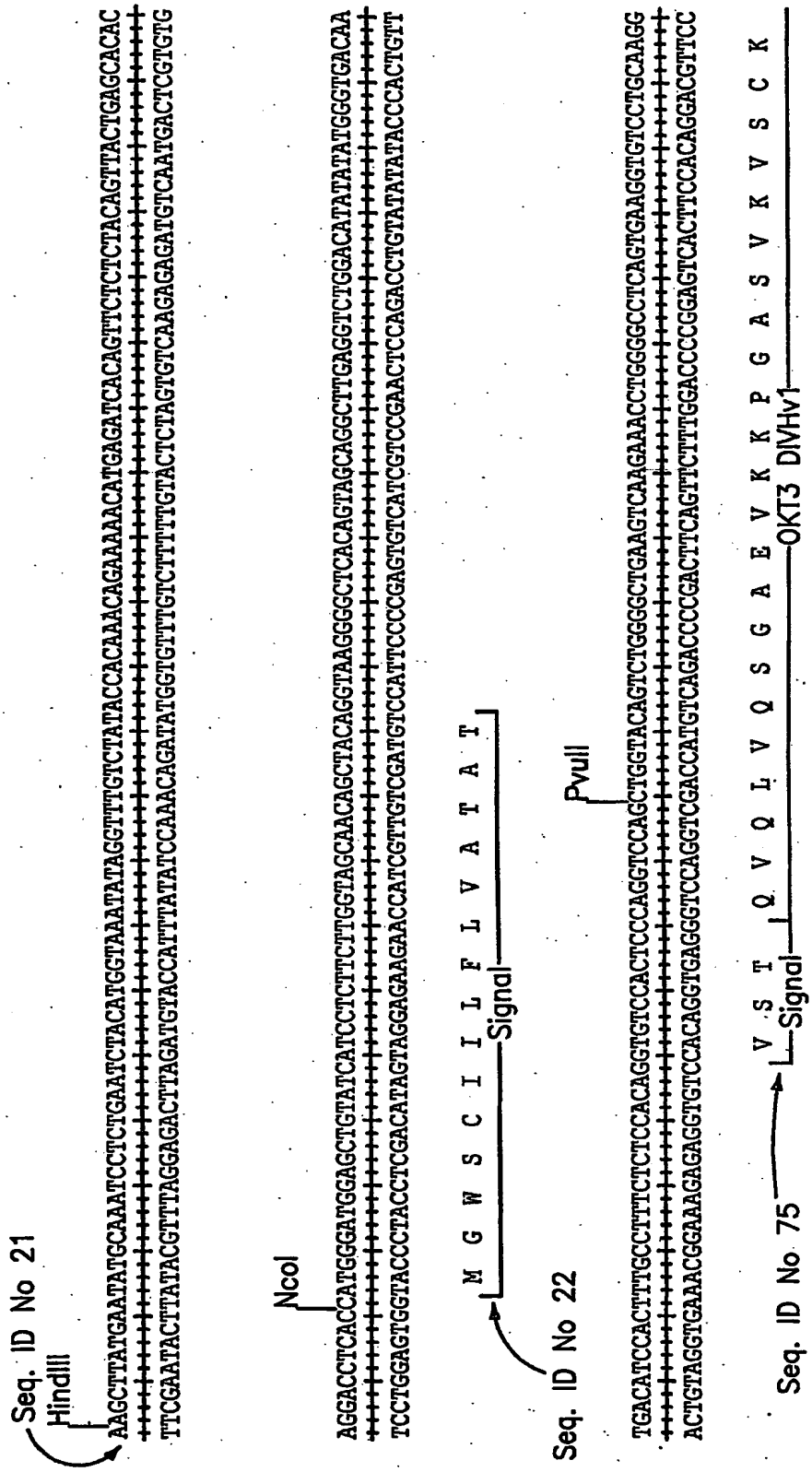
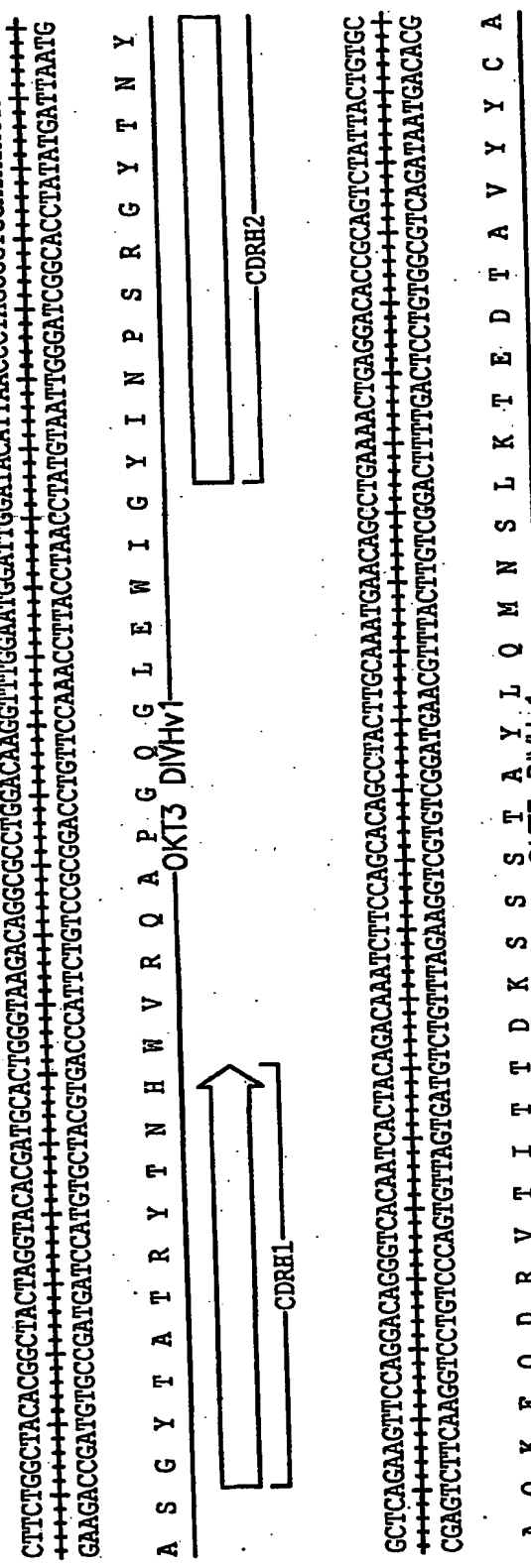


FIG. 13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GAAGACCGATGTGCCGATGATCCATGTGCTACGTGACCCATTCTGTCCGCGGACCTGTTCCAAACCTTACCTAACCTATGTAATTGGGATCGGCACCTATATGATTAATG
A S G Y T A T R Y T N H W V R Q A P G Q G L E W I G Y I N P S R G Y T N Y
OKT3 DIVHv1
CDRH1
CDRH2
GCTCAGAAGTTCCAGGACAGGGTCACAATCACTACAGACAAATCTTCCAGCACAGCCTACTTGCAAATGAACAGCCTGAAAACTGAGGACACCGCAGTCTATTACTGTGC
CGAGTCTTCAAGGTCCTGTCCCAGTGTTAGTGATGTCTGTTTAGAAGGTCGTGTCGGATGAACGTTTACTTGTCGACTTTTGACTCCTGTGGCGTCAGATAATGACACG
A Q K F Q D R V T I T T D K S S S T A Y L Q M N S L K T E D T A V Y Y C A
OKT3 DIVHv1
CDRH2

FIG. 13 (Cont.)

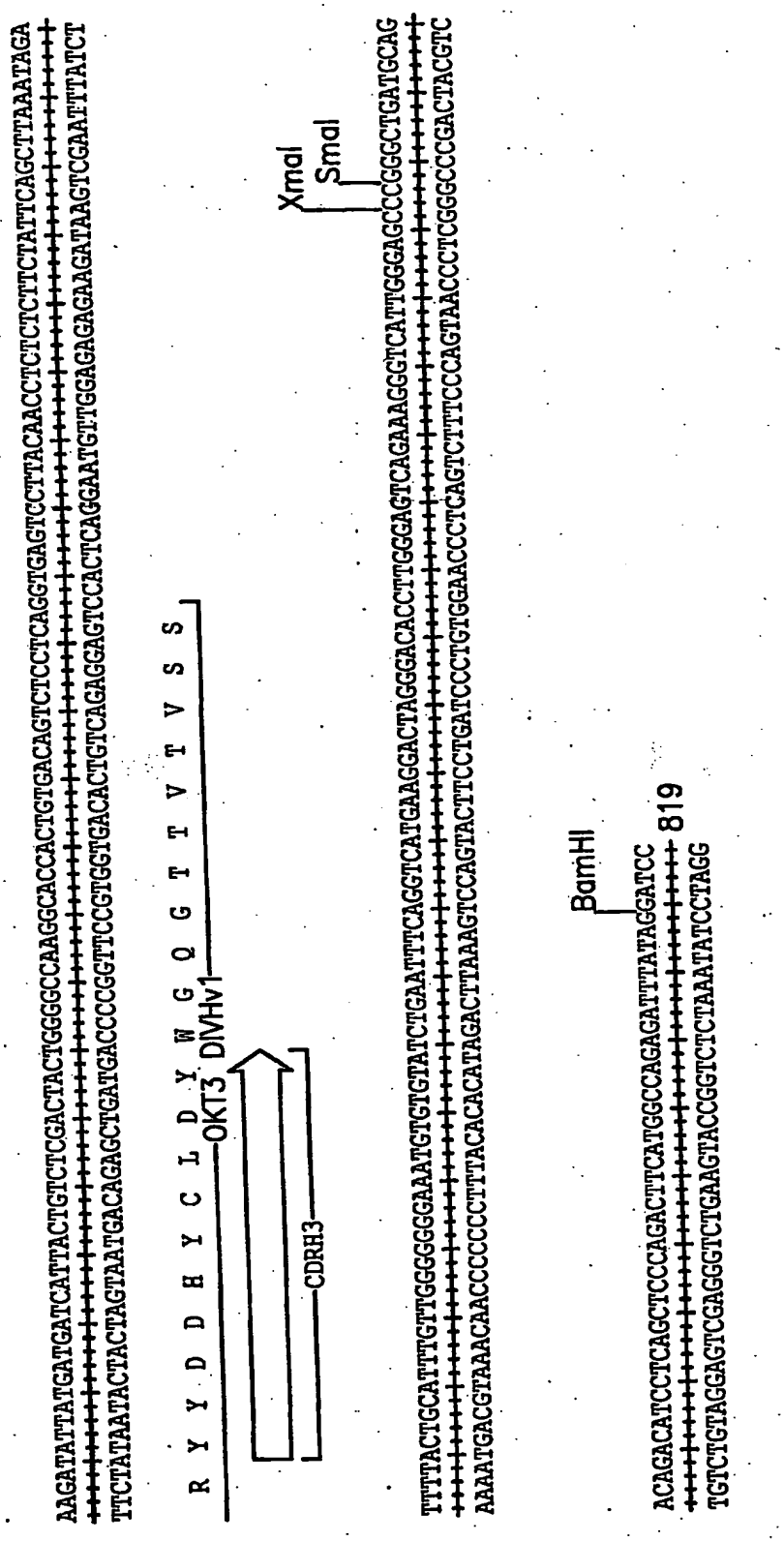
AAGATATTATGATGATCATTACTGTCTCGACTACTGGGGCCAAGGCACCACTGTGACAGTCTCCTCAGGTGAGTCCTTACAACCTCTCTCTTCTATTCAGCTTAAATAGA
TTCTATAATACTACTAGTAATGACAGAGCTGATGACCCCGGTTCCGTGGTGACACTGTCAGAGGAGTCCACTCAGGAATGTTGGAGAGAAGATAAGTCGAATTTATCT
R Y Y D D H Y C L D Y W G Q G T T V T V S S
OKT3 DIVHv1
CDRH3
XmaI
SmaI
TTTTACTGCATTTGTTGGGGGGGAAATGTGTATCTGAATTTCAGGTCATGAAGGACTAGGGACACCTTGGGAGTCAGAAAGGGTCATTGGGAGCCCGGGCTGATGCAG
AAAATGACGTAAACAACCCCCCTTTACACACATAGACTTAAAGTCCAGTACTTCCTGATCCCTGTGGAACCCTCAGTCTTTCCCAGTAACCCTCGGGCCCGACTACGTC
BamHI
ACAGACATCCTCAGCTCCCAGACTTCATGGCCAGAGATTTATAGGATCC 819
TGTCTGTAGGAGTCGAGGGTCTGAAGTACCGGTCTCTAAATATCCTAGG

FIG. 13 (Cont.)

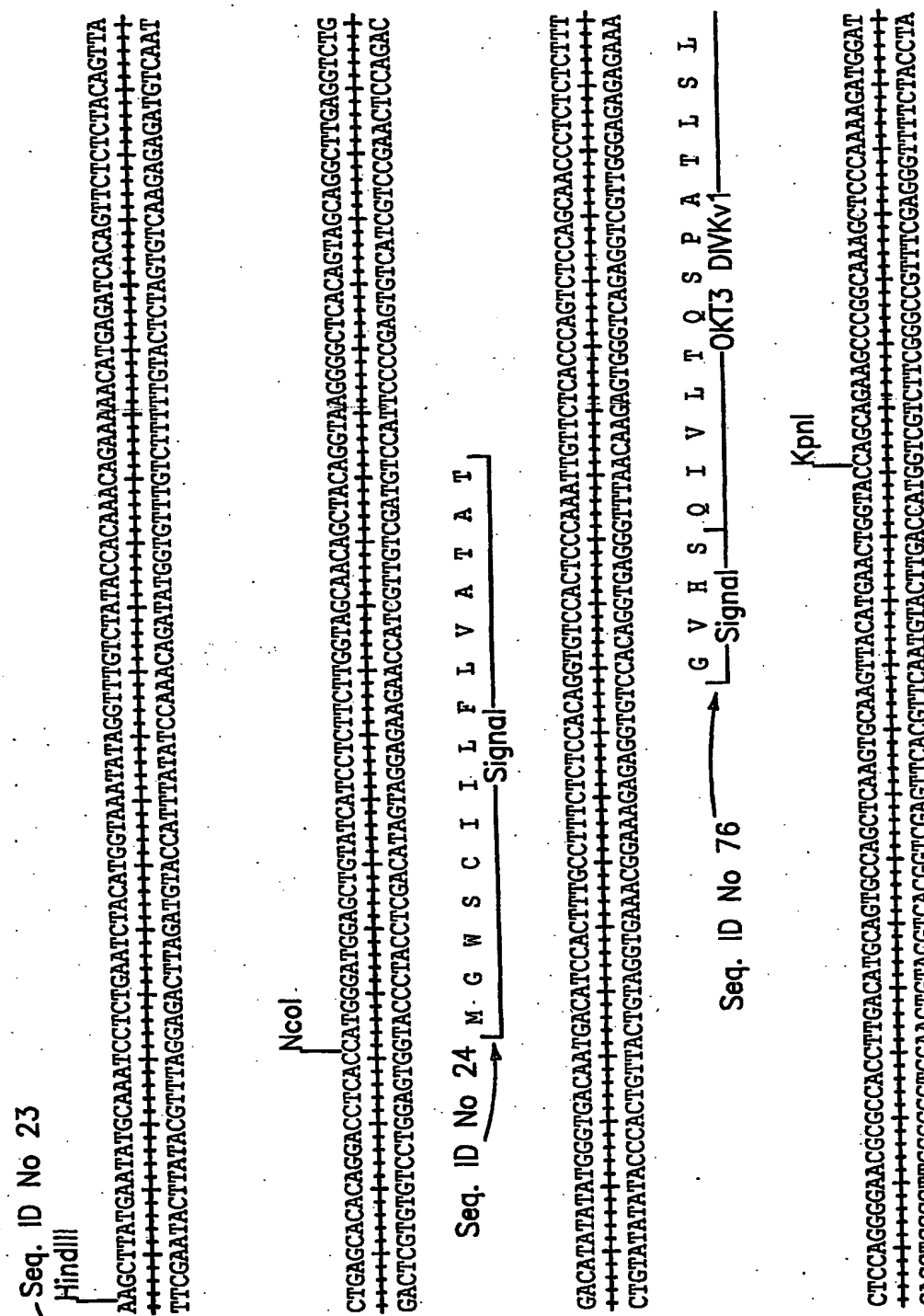
DNA and Amino Acid Sequence of Delmmunised OKT3 VK version 1.
Seq. ID No 23
HindIII
AAGCTTATGAATATGCAAATCCTCTGAATCTACATGGTAAATATAGGTTTGTCTATACCACAAACAGAAAAACATGAGATCACAGTTCTCTACAGTTA
TTCGAATACTTATACGTTTAGGAGACTTAGATGTACCATTTATATCCAAACAGATATGGTGTTTGTCTTTTTGTACTCTAGTGTCAAGAGAGATGTCAAT
NcoI
CTGAGCACACAGGACCTCACCATGGGATGGAGCTGTATCATCCTCTTCTTGGTAGCAACAGCTACAGGTAAGGGGCTCACAGTAGCAGGCTTGAGGTCTG
GACTCGTGTGTCCTGGAGTGGTACCCTACCTCGACATAGTAGGAGAAGAACCATCGTTGTCGATGTCCATTCCCCGAGTGTCATCGTCCGAACTCCAGAC
Seq. ID No 24
M G W S C I I L F L V A T A T
Signal
GACATATATGGGTGACAATGACATCCACTTTGCCTTTCTCTCCACAGGTGTCCACTCCCAAATTGTTCTCACCCAGTCTCCAGCAACCCTCTCTTT
CTGTATATACCCACTGTTACTGTAGGTGAAACGGAAAGAGAGGTGTCCACAGGTGAGGGTTTAACAAGAGTGGGTCAGAGGTCGTTGGGAGAGAAA
Seq. ID No 76
G V H S Q I V L T Q S P A T L S L
Signal
OKT3 DIVKv1
KpnI
CTCCAGGGGAACGCGCCACCTTGACATGCAGTGCCAGCTCAAGTGCAAGTTACATGAACTGGTACCAGCAGAAGCCCGGCAAAGCTCCCAAAAGATGGAT
GAGGTCCCCTTGCGCGGTGGAACTGTACGTCACGGTTCAGTTCACGTTCAATGTACTTGACCATGGTCGTCTTCGGGCCGTTTCGAGGGTTTTCTACCTA

FIG. 14

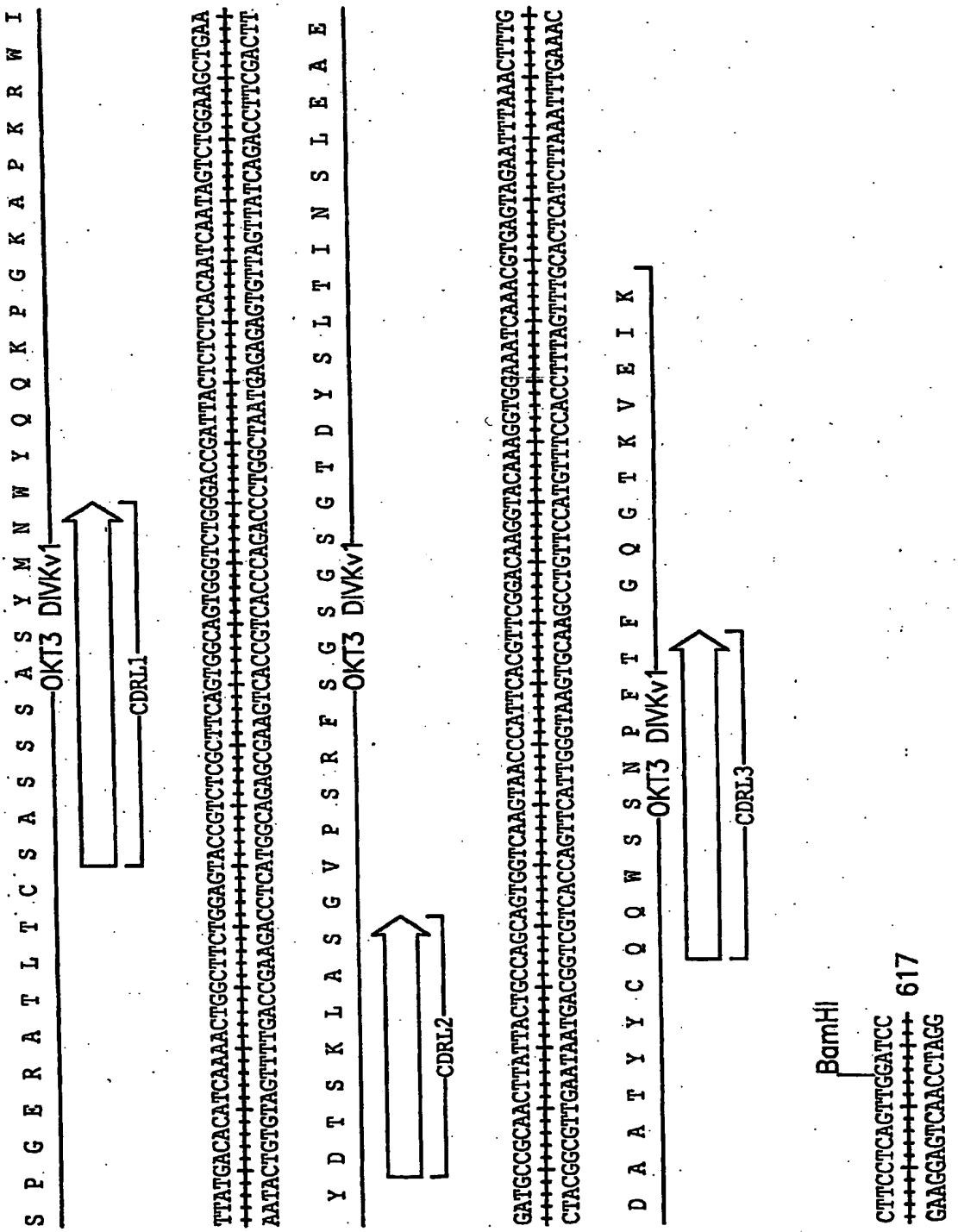
S P G E R A T L T C S A S S S A S Y M N W Y Q Q K P G K A P K R W I
OKT3 DIVKv1
CDRL1
TTATGACACATCAAAACTGGCTTCTGGAGTACCGTCTCGCTTCAGTGGCAGTGGGTCTGGGACCGATTACTCTCTCACAATCAATAGTCTGGAAGCTGAA
AATACTGTGTAGTTTTGACCGAAGACCTCATGGCAGAGCGAAGTCACCGTCACCCAGACCCTGGCTAATGAGAGTGTTAGTTATCAGACCTTCGACTT
Y D T S K L A S G V P S R F S G S G S G T D Y S L T I N S L E A E
OKT3 DIVKv1
CDRL2
GATGCCGCAACTTATTACTGCCAGCAGTGGTCAAGTAACCCATTCACGTTCGGACAAGGTACAAAGGTGGAAATCAAACGTGAGTAGAATTTAAACTTTG
CTACGGCGTTGAATAATGACGGTCGTCACCAGTTCATTGGGTAAGTGCAAGCCTGTTCCATGTTTCCACCTTTAGTTTGCACTCATCTTAAATTTGAAAC
D A A T Y Y C Q Q W S S N P F T F G Q G T K V E I K
OKT3 DIVKv1
CDRL3
BamHI
CTTCCTCAGTTGGATCC
GAAGGAGTCAACCTAGG
617

FIG. 14 (Cont.)

Murine and Chimaeric OKT3 binding to Jurkat, JRT3 and HPB–ALL cells
Values represent the positive % of gated cells in M1

Cell Type	Passage #	Murine OKT3	Mouse Isotype Control	Chimaeric OKT3	Human Isotype Control
Jurkat	12	81.20	0.5	94.68	0.44
JRT3	14	3.45	0.26	4.56	0.43
HPB–ALL	10	99.63	0.62	99.39	0.29

FIG. 15

Antibody	Clone No.	% Cells in M1	
		HPB-ALL	JRT3
Chimaeric OKT3	N/A	99.74	7.74
Control no OKT3 no PE	N/A	2.22	2.3
Control no OKT3 with PE	N/A	2.3	2.21
DMEM Control	N/A	1.91	2.42
DIVH1/DIVK1	19D6	93.87	2.16
DIVH2/DIVK1	24C12	28.47	2.34
DIVH3/DIVK1	27F6	84.75	2.28
DIVH4/DIVK1	30F7	93.06	2.65
DIVH5/DIVK1	35F2	98.15	2.77
DIVH6/DIVK1	37E9	97.85	3.08
DIVH7/DIVK1	42E7	98.62	3.12

FIG. 16

Table 3:

Antibody	Clone No.	% Cells in M1	
		HPB-ALL	JRT3
Chimaeric OKT3	N/A	99.95	0.1
Control no OKT3 no PE	N/A	0.1	0.02
DIVHv1/DIVK2	48G3	20.18	0.1
DIVHv2/DIVK2	52B8	90.04	0.25
DIVHv3/DIVK2	55G5	84.73	0.14
DIVHv4/DIVK2	55B2	69.26	0.13
DIVHv6/DIVK2	66C6	98.16	0.53
DIVHv7/DIVK2	70G10	95.57	0.66

FIG. 17

Competition assay. Inhibition of binding biotinylated mouse OKT3 by chimaeric and DeImmunised OKT3 antibodies, DIVHv1/DIVKv1, DIVHv3/DIVKv1, DIVHv5/DIVKv1, DIVHv6/DIVKv1, OKT3DIVH7/DIVKv1.

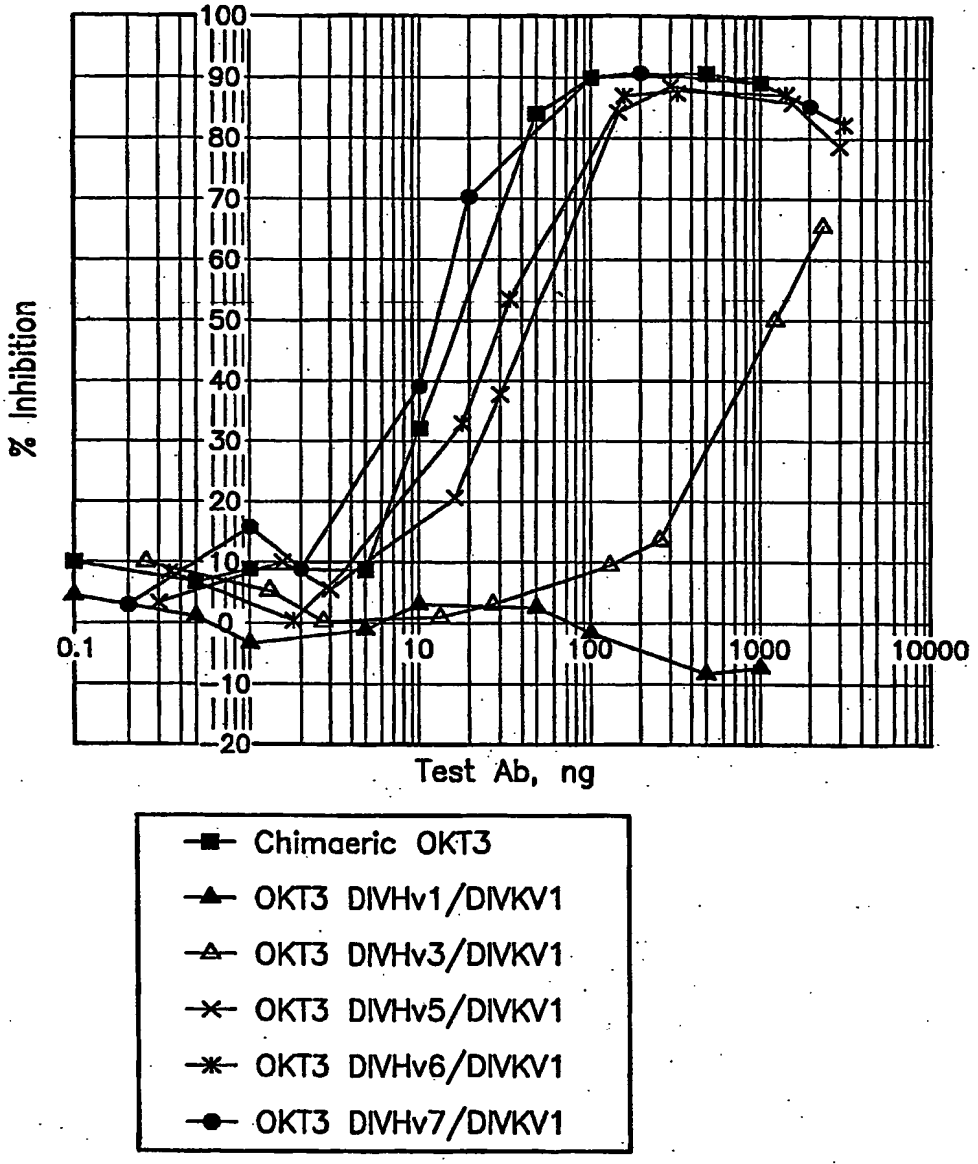

FIG. 18

Competition assay. Inhibition of binding biotinylated mouse OKT3 by mouse, chimaeric and DeImmunised OKT3 antibodies DIVHv2/DIVKv1, DIVHv4/DIVKv1.

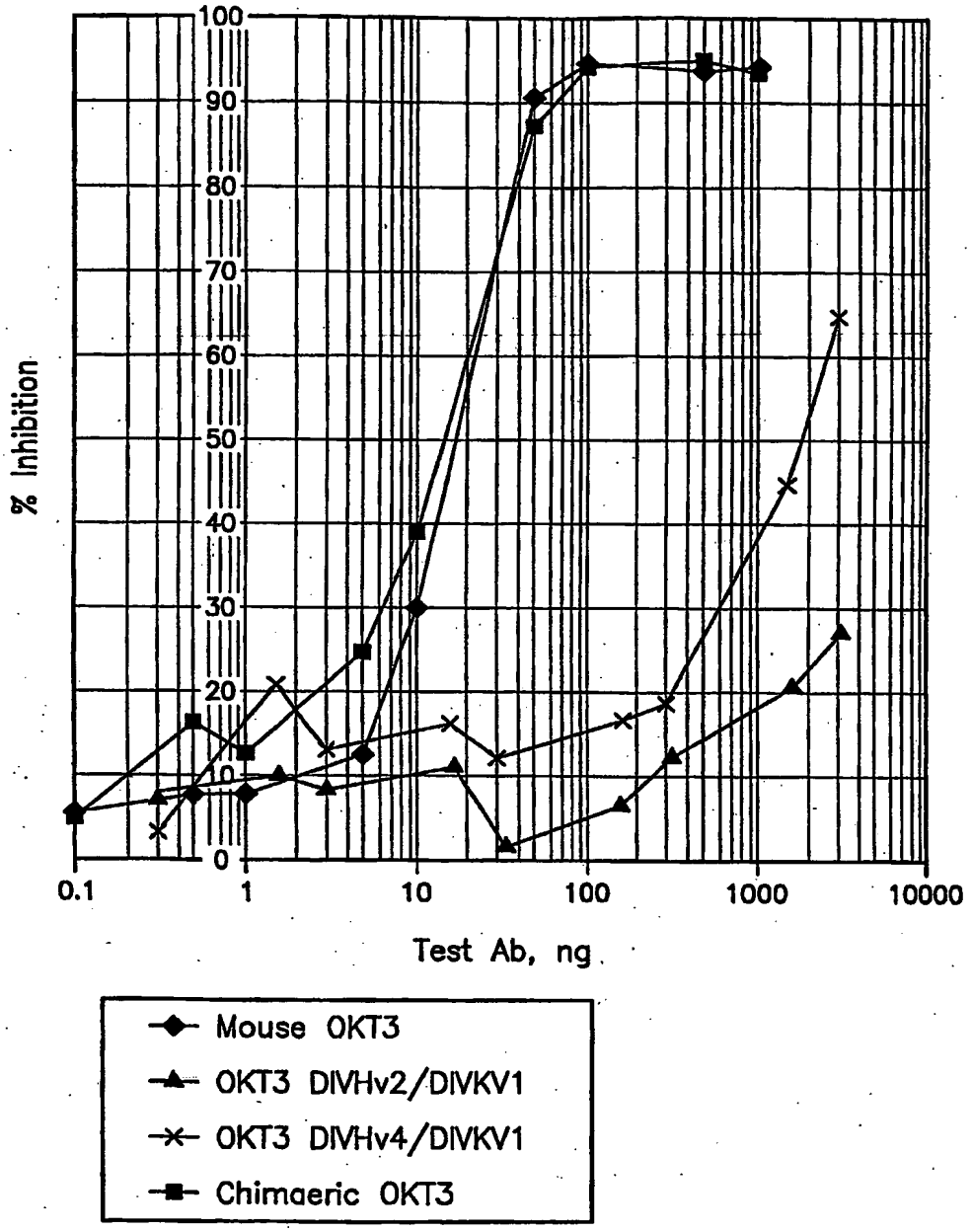

FIG. 19

Competition assay. Inhibition of binding biotinylated mouse OKT3 by mouse, chimaeric and DeImmunised OKT3 antibodies DIVHv3/DIVKv2, DIVHv7/DIVKv2.

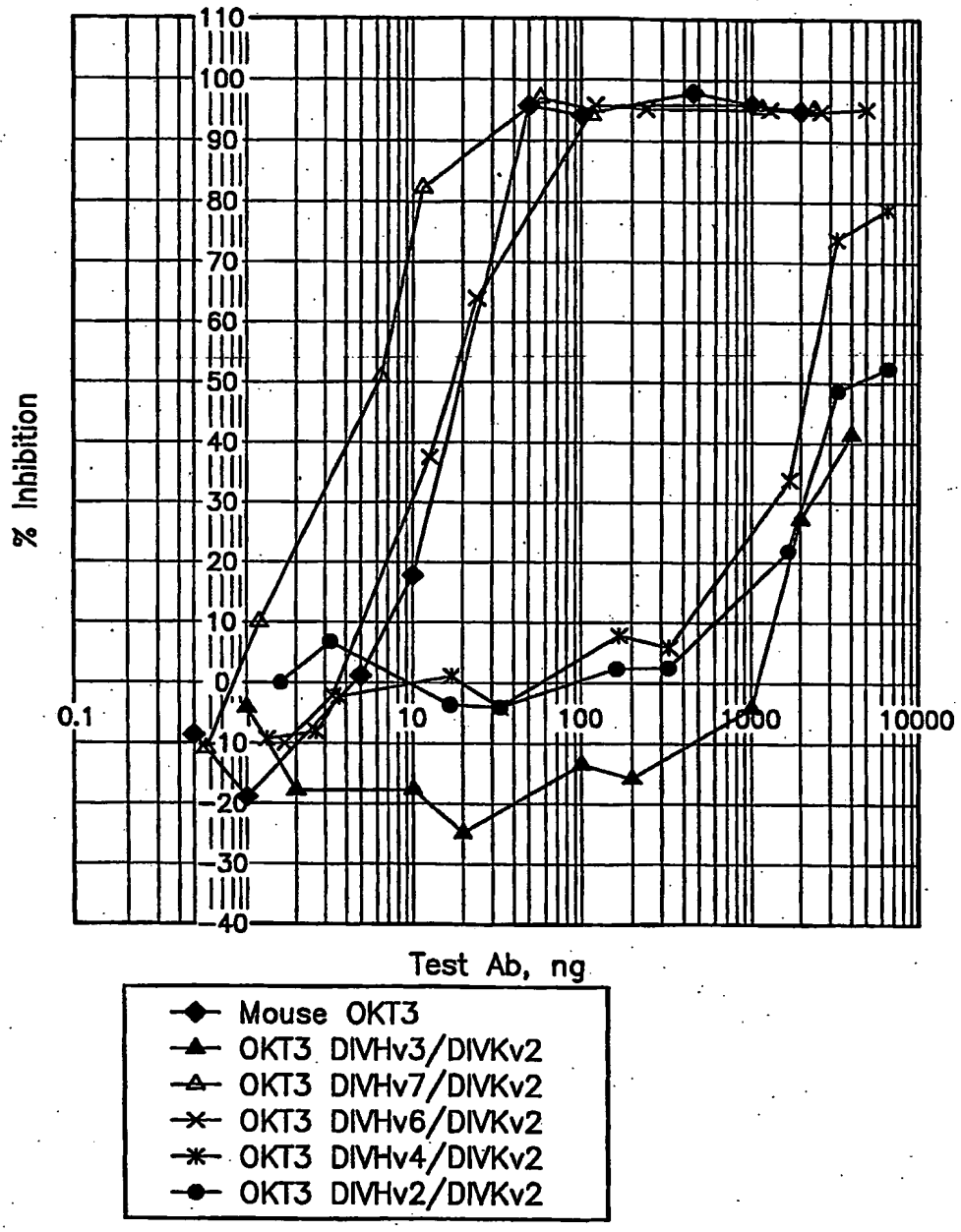

FIG. 20

Competition assay. Inhibition of binding biotinylated mouse OKT3 by mouse, chimaeric and Delmmunised OKT3 antibodies DIVHv1/DIVKv2, DIVHv1/DIVKv1, DIVHv5/DIVKv2, DIVHv5/DIVKv1.

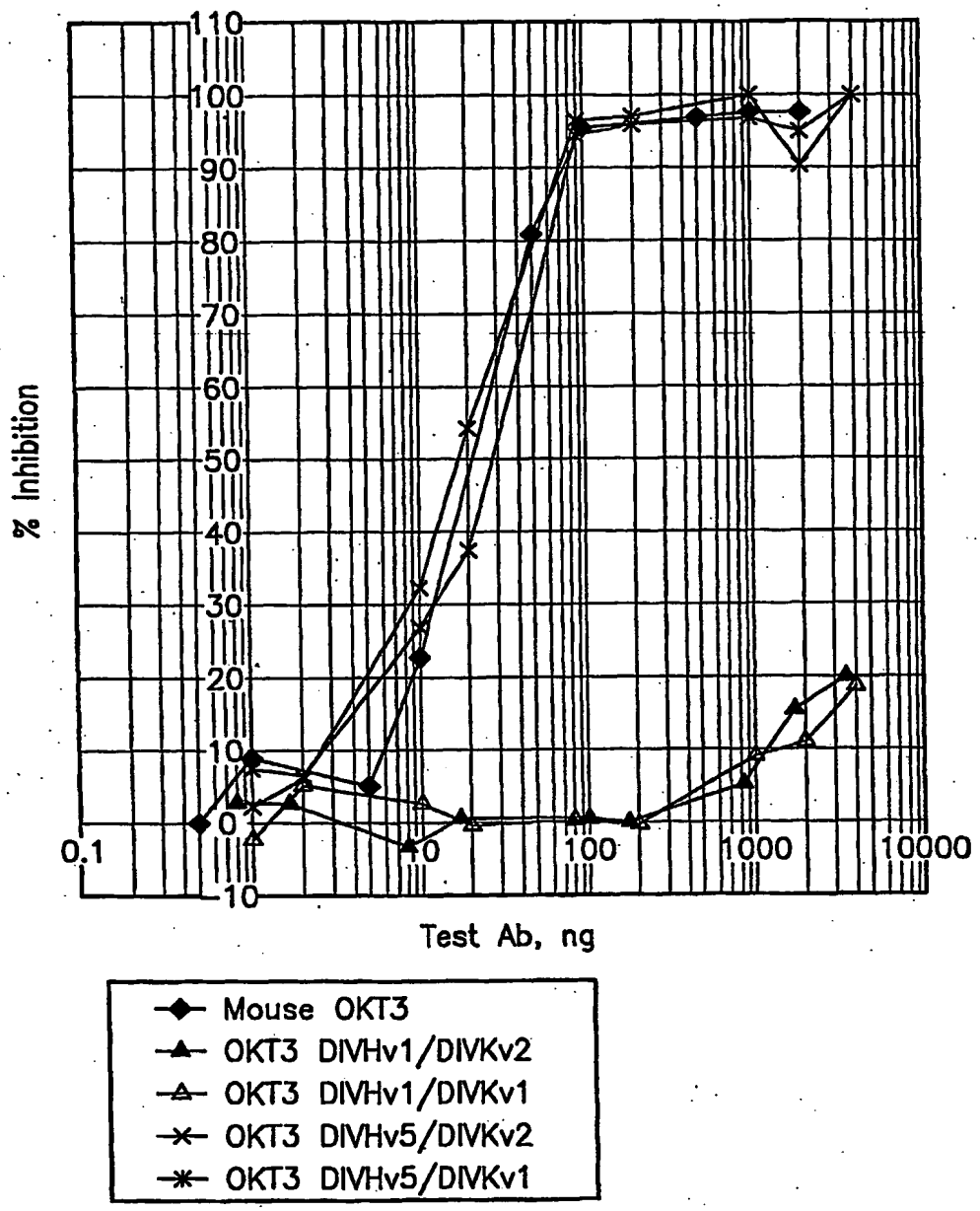

FIG. 21

The IC_{50} determinted from these plots are shown in Table 4.

Table 4:

ANTIBODY	IC50 (ng)
Murine OKT3 1	18
Murine OKT3 2	19
Murine OKT3 3	20
~~Chimeric OKT3 1~~	~~18~~
Chimeric OKT3 2	15
Di-immunized OKT3 DIVHv1/DIVKv1	N/A
Delm OKT3 DIVHv1/DIVKv1 2nd prep	>2000
De-immunized OKT3 DIVHv2/DIVKv1	>3000
De-immunized OKT3 DIVHv3/DIVKv1	1250
De-immunized OKT3 DIVHv4/DIVKv1	1900
De-immunized OKT3 DIVHv5/DIVKv1	45
Delm OKT3 DIVHv5/DIVKv1 2nd prep	19
De-immunized OKT3 DIVHv6/DIVKv1	30
De-immunized OKT3 DIVHv7/DIVKv1	12
De-immunized OKT3 DIVHv1/DIVKv2	>2000
De-immunized OKT3 DIVHv2/DIVKv2	>3000
De-immunized OKT3 DIVHv3/DIVKv2	>4000
De-immunized OKT3 DIVHv4/DIVKv2	2100
De-immunized OKT3 DIVHv5/DIVKv2	28
De-immunized OKT3 DIVHv6/DIVKv2	18
De-immunized OKT3 DIVHv7/DIVKv2	6

FIG. 22